

2002 ACCOMPLISHMENT REPORT

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ANALYTICAL AND NATURAL PRODUCTS CHEMISTRY LABORATORY
CENTER FOR PLANT HEALTH SCIENCE AND TECHNOLOGY
PLANT PROTECTION AND QUARANTINE
U.S. DEPARTMENT OF AGRICULTURE

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These reports were prepared for the information of the U.S. Department of Agriculture, Animal and Plant Health Inspection Service personnel, and others interested in imported fire ant control programs. Statements and observations may be based on preliminary or uncompleted experiments; therefore, the data are not ready for publication or public distribution.

Results of insecticide trials are reported herein. Mention of trade names or proprietary products does not constitute an endorsement or recommendation for use by the U.S. Department of Agriculture.

Compiled and Edited by:

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April 2003

2002 IMPORTED FIRE ANT OBJECTIVES

GULFPORT PLANT PROTECTION STATION GULFPORT, MS

OBJECTIVE 1: Development and refinement of quarantine treatments for certification of regulated articles.

- Emphasize development of quarantine treatments for containerized nursery stock.
- Evaluate candidate toxicants, formulation, and dose rates for various use patterns.
- Test and evaluate candidate pesticides for use on grass sod and field grown nursery stock.
- Assist in registration of all treatments shown to be effective.

OBJECTIVE 2: Advancement of technology for population suppression and control.

- New product/formulation testing and evaluation.
- Conduct label expansion studies.
- Evaluation of non-chemical biocontrol agents, including microbial, nematodes, and predaceous arthropods.

OBJECTIVE 3: Preparation/distribution of technical information on control, quarantine procedures, new technology, biological hazards, etc., to state agencies, the media, and the public.

- Provide training to state regulatory agencies and nursery associations.
- Publish and distribute informational aids for state agencies, nursery associations, PPQ personnel, and other interested stakeholders.

OBJECTIVE 4: Determine impact of IFA on biodiversity of various ecosystems.

- Provide technical support and assistance to other research organizations such as ARS, Universities, Mississippi Nature Conservancy, etc. to expedite ecological studies on the impact of IFA on T&E species.
- Conduct bait transects and compare current myrmecofaunal records with similar surveys done in the past to determine impact of IFA on other ant species.

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PROJECT NO: A9P01

PROJECT TITLE: Residual Activity of Various Drench Treatments in Nursery Potting Media,
2002

TYPE REPORT: Final

LEADER/PARTICIPANTS: Lee McAnally

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin or tefluthrin) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping by smaller nurseries which ship limited quantities of plant material outside the quarantine area, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement

Drench trials are a part of the ongoing search for new and/or better toxicants for use in the IFA quarantine program.

METHODS AND MATERIALS:

Trade 1 gallon nursery containers were filled with potting media (MAFES mix – 3:1:1 mix of pine bark, peat moss, and sand (bulk density 850# per cu. yd.). Containers with media were allowed to sit for 5-7 days in a simulated nursery area with overhead irrigation applied at a rate of approximately 1.5 inches water per week (irrigation system runs nightly). On February 5, 2002, the containerized media was treated with the candidate treatments. 400 ml of each drench solution (approximately 1/5 volume of container) was applied to each container. Control containers were drenched with 400 ml plain water. Standard alate female bioassays (Appendix I) were performed at 1 week, 2 weeks, 1 month, and monthly thereafter.

Rates of application were as follows:

Talstar® F	bifenthrin	25 ppm	3.55ml / gal H ₂ O
		50 ppm	7.1ml / gal H ₂ O
Scimitar® SC	lambda-cyhalothrin	25 ppm	2.6ml / gal H ₂ O
		50 ppm	5.2ml / gal H ₂ O

DeltaGard®	deltamethrin	25 ppm	5.7ml / gal H ₂ O
		50 ppm	11.4ml / gal H ₂ O
Demon®	cypermethrin	50 ppm	2.34ml / gal H ₂ O
		100 ppm	4.68ml / gal H ₂ O

RESULTS:

Results are summarized in Table 1. Through the termination of the test (6 months) all formulations except Demon attained 100% mortality within 5 days post-exposure to treated media. Demon maintained 100% mortality through 2 months at the 100 ppm rate and through 1 month at the 50 ppm rate.

Table 1. Residual Activity of Various Drench Treatments in Nursery Potting

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		1 wk	2 wks	1	2	3	4	5	6
Talstar	25	100(1)	100(1)	100(1)	100(3)	100(3)	100(3)	100(3)	100(3)
	50	100(1)	100(1)	100(1)	100(3)	100(3)	100(3)	100(3)	100(3)
Scimitar	25	100(1)	100(1)	100(1)	100(3)	100(3)	100(4)	100(3)	100(3)
	50	100(1)	100(1)	100(1)	100(3)	100(3)	100(4)	100(3)	100(3)
Deltagard	25	100(1)	100(1)	100(1)	100(3)	100(4)	100(5)	100(3)	100(4)
	50	100(1)	100(1)	100(1)	100(3)	100(3)	100(3)	100(3)	100(3)
Demon	50	100(1)	100(1)	100(1)	75	45	75	15	10
	100	100(1)	100(1)	100(1)	100(12)	85	85	50	35
Check	---	0	0	0	10	10	15	5	10

PROJECT NO: A9P01 - GPPS00-01

PROJECT TITLE: Further Testing of Chlorfenapyr as an Imported Fire Ant Quarantine Treatment (2000)

REPORT TYPE: Interim

PROJECT LEADER/PARTICIPANT(s): Lee McAnally

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin or tefluthrin) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement.

Chlorfenapyr is an experimental insecticide-miticide under development by American Cyanamid (Princeton, NJ). The product is active against many pests, and works as a broad spectrum contact and stomach poison. Previously we tested a liquid formulation to determine whether the product showed significant activity against IFA in containerized nursery stock. In August 1997, we began testing a 0.5G granular formulation as an incorporated treatment (FA01G097).

In August 1999, we initiated an expanded test of chlorfenapyr using a 2SC liquid formulation as a drench treatment, as well as 1G, 1.5G and 2G formulations each on two different carriers (clay and corn cob grit) as incorporated treatments. All of these treatments were applied to three different potting media (FA01G019).

In August 2000, another trial was initiated using the 1G and 1.5G formulations on the grit carrier.

MATERIALS AND METHODS:

Incorporated Treatments:

Granular treatments included 1% and 1.5% products formulated on a corn cob grit carrier. Each of the granular formulations was blended into the MAFES media (3:1:1 pine bark: sphagnum peat moss: sand - bulk density = 785 lb/cu yd) at rates of 50, 75, 100 and 200 ppm. A portable cement mixer (2 cu ft capacity) was used to blend the toxicant into the potting media, and was

operated for 15 minutes per batch to insure thorough blending. Treated media was then poured into one-gallon capacity plastic nursery pots and weathered outdoors under simulated nursery conditions. A pulsating overhead irrigation system supplied ca. 1-1½ inches water per week. At monthly intervals, subsamples were taken from 3 pots of each treatment and composited and subjected to standard alate queen bioassay. The 1.0G formulation was mixed on August 28 and the 1.5G formulation was mixed on August 29, 2000.

RESULTS:

All rates are producing 100% mortality in 12 days exposure or less through 27 months post-treatment (Table 1), showing excellent potential as a preplant incorporation treatment for the IFA quarantine. Additional trials in various media types would need to be completed to ensure efficacy does not decrease in other media types. Discussion with the company will precede any future testing and current trial will run through 30 months and then be terminated.

Table 1. Residual activity of chlorfenapyr 1.0G and 1.5G.

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		1	2	3	5	6	7	8	9
1.0G	50	100(6)	100(8)	100(11)	100(10)	100(10)	100(8)	100(13)	100(8)
	75	100(5)	100(7)	100(7)	100(9)	100(7)	100(7)	100(6)	100(7)
	100	100(4)	100(7)	100(8)	100(9)	100(6)	100(7)	100(7)	100(7)
	200	100(3)	100(7)	100(5)	100(6)	100(4)	100(3)	100(8)	100(6)
1.5G	50	100(6)	100(8)	100(11)	100(11)	100(10)	100(9)	100(7)	100(9)
	75	100(6)	100(8)	100(8)	100(11)	100(7)	100(7)	100(7)	100(7)
	100	100(6)	100(4)	100(7)	100(9)	100(7)	100(7)	100(6)	100(7)
	200	100(4)	100(4)	100(5)	100(6)	100(6)	100(7)	100(6)	100(7)
	Check*	15	10	5	5	5	5	30	5

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		10	11	12	13	14	15	17	18
1.0G	50	100(10)	100(6)	100(9)	100(11)	100(12)	100(7)	100(9)	100(10)
	75	100(10)	100(6)	100(8)	100(11)	100(11)	100(7)	100(6)	100(6)
	100	100(7)	100(6)	100(7)	100(8)	100(7)	100(7)	100(6)	100(6)
	200	100(7)	100(4)	100(7)	100(6)	100(5)	100(4)	100(6)	100(4)
1.5G	50	100(10)	100(6)	100(8)	100(8)	100(11)	100(8)	100(8)	100(7)
	75	100(7)	100(4)	100(7)	100(7)	100(7)	100(7)	100(6)	100(7)
	100	100(6)	100(6)	100(7)	100(7)	100(6)	100(7)	100(6)	100(5)
	200	100(10)	100(3)	100(6)	100(5)	100(5)	100(4)	100(6)	100(4)
	Check*	10	10	5	10	5	0	0	0

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		19	20	21	22	23	24	25	26
1.0G	50	100(11)	100(11)	100(10)	100(10)	100(7)	100(7)	100(11)	100(10)
	75	100(10)	100(10)	100(7)	100(7)	100(7)	100(6)	100(8)	100(7)
	100	100(6)	100(6)	100(5)	100(5)	100(6)	100(6)	100(8)	100(6)
	200	100(6)	100(6)	100(5)	100(5)	100(5)	100(6)	100(4)	100(5)
1.5G	50	100(11)	100(10)	100(10)	100(7)	100(7)	100(6)	100(9)	100(7)
	75	100(10)	100(10)	100(7)	100(6)	100(6)	100(6)	100(8)	100(7)
	100	100(6)	100(6)	100(7)	100(5)	100(6)	100(6)	100(8)	100(5)
	200	100(5)	100(5)	100(5)	100(4)	100(4)	100(2)	100(4)	100(5)
	Check*	5	0	10	5	10	10	5	10

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		27							
1.0G	50	100(11)							
	75	100(10)							
	100	100(6)							
	200	100(5)							
1.5G	50	100(10)							
	75	100(11)							
	100	100(11)							
	200	100(11)							
	Check*	10							

*Check mortality is shown at longest exposure time

PROJECT NO: A9P01

PROJECT TITLE: Residual Activity of TopPro Specialties' Formulation of Bifenthrin, 2002

REPORT TYPE: Interim

PROJECT LEADER/PARTICIPANTS: Lee McAnally, Shannon James

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery stock include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin or tefluthrin) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement.

TopPro Specialties, Micro Flo Company has begun the manufacture of bifenthrin in both granular (0.2%) and liquid flowable (7.9%) formulations. The granular formulation is produced on two different carriers, Sand and DG Lite. In August 2002 a study was initiated to determine the efficacy of TopPro bifenthrin. Each formulation was set up in treatment rates equivalent to those specified in the quarantine treatment manual for durations corresponding to the certification periods for each treatment rate.

MATERIALS AND METHODS:

Granular Incorporation Treatment:

Both formulations (carriers) of granular TopPro were blended into the MAFES media (3:1:1 pine bark: sphagnum peat moss: sand - bulk density = 850 lb/cu yd) at rates of 10, 12, 15, and 25 ppm. A portable cement mixer (2 cu ft capacity) was used to blend the toxicant into the potting media, and was operated for 15 minutes per batch to insure thorough blending. Treated media was then poured into one-gallon capacity plastic nursery pots and weathered outdoors under simulated nursery conditions. A pulsating overhead irrigation system supplied ca. 1-1½ inches water per week. At monthly intervals, sub samples were taken from 2 pots of each treatment and composited and subjected to standard alate queen bioassay (Appendix I).

Drench Treatment:

Untreated media was placed in 1-gallon nursery pots and drenched with 400ml finished solution at a rate of 25 ppm. The pots were then placed under the same conditions and tested in the manner described above.

RESULTS:

Through 5 months post-treatment all rates and formulations have produced 100% mortality in 3 days or less. Results are summarized in Table 1. The TopPro Specialties flowable bifenthrin formulation in the MAFES media is as effective as the FMC formulation indicating acceptability as a product to be used in the IFA quarantine. To ensure efficacy across media types, additional testing in other media types should be initiated. The granular testing will continue for a minimum of 24 months, or until efficacy decreases, and additional testing of this formulation should also occur in other media types.

Table 1. Residual activity of TopPro bifenthrin.

Formulation Tested	Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)							
		1	2	3	4	5	6		
DG lite carrier	10	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	12	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	15	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	25	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
Sand carrier	10	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	12	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	15	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	25	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
Drench	25	100(1)	100(3)	100(1)	100(3)	100(3)	100(1)		
	Check	0	0.	0	0	0	0		

PROJECT NO: A9P01

PROJECT TITLE: Residual Activity of Granular Deltagard G Incorporated into Nursery Potting Media, 2002

REPORT TYPE: Interim

PROJECT LEADER/PARTICIPANTS: Shannon Wade, Lee McAnally

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin, tefluthrin or fipronil) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement. However, actual products with labels for use in the IFA quarantine are limited, therefore we regularly screen potential insecticides for these use patterns.

Bayer Corp. has developed Deltagard 0.1G for testing as a granular incorporation treatment. On May 21, 2002, we initiated a test to determine residual activity as a soil incorporation treatment.

MATERIALS AND METHODS:

The formulation was blended into the MAFES media (3:1:1 pine bark: sphagnum peat moss: sand - bulk density = 850 lb/cu yd) at rates of 25, 50, and 75 ppm. A portable cement mixer (2 cu ft capacity) was used to blend the toxicant into the potting media, and was operated for 15 minutes per batch to insure thorough blending. Treated media was then poured into one-gallon capacity plastic nursery pots and weathered outdoors under simulated nursery conditions. A pulsating overhead irrigation system supplied ca. 1-1½ inches water per week. At monthly intervals, sub-samples were taken from 3 pots of each treatment and composited and subjected to standard alate queen bioassay (Appendix I).

RESULTS:

All rates are producing 100% mortality in 12 days exposure or less through 10 months post-treatment (Table 1). Evaluations will continue until all rates lose efficacy.

Table 1. Residual Activity of Deltagard G as an Incorporated Treatment

Rate of Application (ppm)	Mean % mortality to alate females at indicated post-treatment interval								
	1 month	2 months	3 months	4 months	5 months	7 months	8 months	9 months	10 months
25	100	100	100	100	100	100	100	100	100
50	100	100	100	100	100	100	100	100	100
75	100	100	100	100	100	100	100	100	100
check	10	0	0	5	20	5	0	5	5

PROJECT NO: A9P01

PROJECT TITLE: Residual Activity of Fluvalinate 0.2 % (Mavrik) as a Granular Incorporated Treatment for Nursery Potting Media, 2001

REPORT TYPE: Final

PROJECT LEADER/PARTICIPANTS: Lee McAnally

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery stock include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin or tefluthrin) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement.

Mavrik is granular synthetic pyrethroid insecticide consisting of fluvalinate 0.2% and piperonyl butoxide 0.4% produced by Wellmark International.

MATERIALS AND METHODS:

On September 7, 2001 granular Mavrik was blended into the MAFES media (3:1:1 pine bark: sphagnum peat moss: sand - bulk density = 800 lb/cu yd) at rates of 10, 25, 50, and 100 ppm. A portable cement mixer (2 cu ft capacity) was used to blend the toxicant into the potting media, and was operated for 15 minutes per batch to insure thorough blending. Treated media was then poured into one-gallon capacity plastic nursery pots and weathered outdoors under simulated nursery conditions. A pulsating overhead irrigation system supplied ca. 1-1½ inches water per week. At monthly intervals, sub samples were taken from 3 pots of each treatment and composited and subjected to standard alate queen bioassay (Appendix I).

RESULTS:

Results are summarized in Table 1. The 10 and 25ppm rates never achieved 100% mortality and were dropped from the test after 9 months. The 50ppm rate maintained 100% mortality through 5 months and began to decline erratically after that. The 100ppm rate also maintained 100% through 5 months and became erratic although it still attained 100% several times through 13

months post-treatment. After 13 months the 100 ppm rate began to decline and the test was terminated.

In 1992-1993, another Mavrik formulation (0.15G) was tested as a granular preplant incorporation treatment with limited results: rates <100 ppm provided <1 month residual activity, 100 ppm provided erratic activity for 1-6 months, and rates >150 ppm effective for 6 months. The current trial confirms those previous results with 100 ppm providing only 5 months of 100% efficacy. Due to the high rate of application needed to achieve 6 months of residual activity, it is probably not economical or practical to pursue this product for use in this IFA quarantine use pattern.

Table 1. Residual Activity of Mavrik 0.2G as a Granular Incorporated Treatment

Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)								
	1	2	3	4	5	6	7	8	
10	50	35	60	30	35	0	55	40	
25	95	70	50	85	70	75	30	55	
50	100(2)	100(9)	60	100(7)	100(11)	80	75	90	
100	100(6)	100(13)	100(11)	100(2)	100(4)	95	100(3)	100(10)	
Check	20	0	5	0	5	0	5	25	

Table 1 (cont.). Residual Activity of Mavrik 0.2G as a Granular Incorporated Treatment

Rate of Application (ppm)	Mean % mortality to alate females at indicated months post-treatment (days required to reach 100% mortality)								
	9	10	11	12	13	14	15		
10	5	*	*	*	*	*	*		
25	40	*	*	*	*	*	*		
50	25	60	30	70	55	40	35		
100	80	100(7)	35	100(10)	100(14)	90	75		
Check	5	10	10	25	5	10	10		

PROJECT NO: A9P01

PROJECT TITLE: Residual Activity of Purcell 4.0% Granular Acephate Incorporated into Nursery Potting Media, 2002

REPORT TYPE: Final

PROJECT LEADER/PARTICIPANTS: Lee McAnally, Shannon Wade

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for containerized nursery include the use of granular insecticides incorporated into potting media or liquid drenches applied prior to shipping. Nursery stock treated with incorporated insecticides (bifenthrin or tefluthrin) may be certified for 6 months to 2 years, depending on the rate incorporated into the media (10-25 ppm based on bulk density of media). This allows the grower to use less insecticide on nursery stock that will be held on site for a short period of time, and more on those that need a longer growing period prior to selling. Drench treatments (chlorpyrifos, diazinon or bifenthrin) are generally used just prior to shipping, and those currently approved for use in the quarantine have certification periods of 10 days to 6 months. Since drench treatments are used just prior to shipping, long residual activity is not a requirement.

Purcell has developed a 4.0% time-release granular acephate for testing as a granular incorporation treatment.

On October 30, 2002, we initiated a test to determine residual activity as a soil incorporation treatment.

MATERIALS AND METHODS:

The formulation was blended into the MAFES media (3:1:1 pine bark: sphagnum peat moss: sand - bulk density = 850 lb/cu yd) at rates of 25, 50, 100 and 200 ppm. A portable cement mixer (2 cu ft capacity) was used to blend the toxicant into the potting media, and was operated for 15 minutes per batch to insure thorough blending. Treated media was then poured into one-gallon capacity plastic nursery pots and weathered outdoors under simulated nursery conditions. A pulsating overhead irrigation system supplied ca. 1-1½ inches water per week. At monthly intervals, sub-samples were taken from 3 pots of each treatment and composited and subjected to standard alate queen bioassay (Appendix I).

RESULTS:

Results through 2 months post-treatment show only 50% or less mortality, therefore due to the ineffectiveness of this formulation, the trial was terminated (Table 1).

Table 1. Residual Activity of Purcell Acephate as an Incorporated Treatment

Rate of Application (ppm)	Mean % mortality to alate females at indicated post-treatment interval		
	2 weeks	4 weeks	2 months
25	20	0	0
50	40	25	10
100	50	0	15
200	25	25	25
check	25	0	10

PROJECT NO: A9P01 - FA01G069

PROJECT TITLE: Effectiveness of Permethrin Impregnated Nursery Pots in Preventing
Imported Fire Ant Invasion of Containerized Nursery Stock, 1999

TYPE REPORT: Final

PROJECT LEADERS: Homer Collins, Anne-Marie Callcott and Shannon Wade

COOPERATORS: Premium Compounded Products, LLC (Corinne Brothers)
Nursery Supplies, Inc. (Henry Guarriello, Jr.)
AgrEvo Environmental Health - now Bayer Environmental Science (John
Lucas/Jing Zhai)
Windmill Nursery (Tom Cooper)

INTRODUCTION:

Nursery stock and other regulated articles cannot be shipped outside the imported fire ant (IFA) quarantined area unless treated with an approved insecticide (7CFR §301.81) to prevent inadvertent spread of IFA. Several treatment options are approved and registered for this use pattern. Both liquid drenches (chlorpyrifos, diazinon, and bifenthrin), and granular insecticides (tefluthrin and bifenthrin) are approved for use. The most frequently used treatment is incorporation of either granular tefluthrin or bifenthrin into the potting media prior to "potting up". The residual activity of the insecticide prevents IFA invasion of containerized nursery stock for up to 24 months, depending upon dose rate employed.

New technologies utilizing insecticides applied to the nursery pot or insecticides impregnated into the plastic of the nursery pot to prevent IFA invasion have been investigated by our laboratory over the past several years. Preliminary work with permethrin impregnated nursery pots has shown the potential for preventing IFA infestation of small nursery containers (report FA01G038). This trial was initiated to expand on our preliminary observations and test the impregnated containers in actual nursery conditions with plants added.

MATERIALS AND METHODS:

Three sizes of nursery containers (1, 3, and 10 gallon) impregnated with permethrin or deltamethrin were produced the week of December 14, 1998 by Premium Compounded Products. Concentrations of permethrin in the plastic were 0.25, 0.50, 0.75 and 1.0%, and of deltamethrin were 0.025, 0.050, 0.075 and 0.10%. Containers were potted up at Windmill Nursery on May 1, 1999. Due to logistics and resources, only three treatments were subjected to bioassay at our laboratory: 0.5 and 1.0% permethrin, and 0.10% deltamethrin. Pots were transported to the Gulfport laboratory quarterly for bioassay testing. The 0.05% deltamethrin concentration was tested at another laboratory at 6 month intervals. The other rates will be held for testing as needed. Other trials, not reported here, were initiated in other nurseries and bioassays performed by other laboratories.

Bioassays were conducted in the laboratory in 2' x 8' test arenas (Figure 1). Sides of the test arena were talced to prevent ants from climbing out and escaping. An impregnated pot was placed at one end of the arena, and an untreated check container filled with potting media was placed at the distal end of the arena. A field collected IFA colony complete with associated soil and nest tumulus was then placed in the center of the arena. Overhead incandescent light bulbs (60 watts, placed 14" above the test arena) slowly desiccated the nest so that the ants were encouraged to migrate to the more moist containers. Therefore, the IFA colony had an equal opportunity to move into either a permethrin pot or the untreated check pot. Pots were observed at 24 hour intervals for 7 days after introduction, and the estimated number of worker ants successfully invading each pot was recorded. A pot was considered infested if there were +25 workers inside the pot. There were 3 replicates per sampling interval.

RESULTS:

Through 16 months after potting up, the 1.0% permethrin impregnated nursery containers excluded IFA in all container sizes (Figs. 2, 3 and 4). However, at 18 months, one 1.0% permethrin 1-gallon container had 50-100 workers in the container, and at 21 and 24 months after treatment one container at each month had ca. 50 workers. Results with both 0.5% permethrin and 0.1% deltamethrin have been erratic. The 3-gallon containers have been the most erratic with these treatments, with 50 to 5000 (whole colony) infesting the treated pots. In conversations with AgrEvo (now Aventis), erratic results have been obtained in most of the trials initiated under this protocol, indicating possible formulation/production problems with this container size.

At 21 months, evaluations of the 0.75% permethrin containers was initiated to determine if this rate was as effective as the 1.0% rate. At 21 months, one 1-gallon, all three 3-gallon, and one 10-gallon container contained 25-100 worker ants (Figures 2, 3 & 4). At 24 months, two 1-gallon and one 3-gallon container contained 100 workers, each.

We received containers from both Simpson Nursery in Monticello, FL and McCorkle Nursery in Dearing, GA for evaluation at ca. 36 months after trial initiation at those nurseries. From Simpson Nursery we received and tested:

1 gallon	3 @ 0.75 permethrin; 3 @ 1.0 permethrin
3 gallon	3 @ 1.0 permethrin (others sent were incorrect rate)
10 gallon	6 @ 0.75 permethrin; 3 @ 1.0 permethrin

From McCorkle Nursery we received and tested:

1 gallon	6 @ 0.75 permethrin; 3 @ 1.0 permethrin
3 gallon	6 @ 0.75 permethrin; 3 @ 1.0 permethrin
10 gallon	none

DISCUSSION:

Data from similar trials initiated at other sites need to be compiled to determine true effectiveness of permethrin impregnated nursery pots at excluding IFA. Results from this trial

indicate that while only a few containers treated with rates of <1.0% permethrin were infested by whole colonies, these containers were regularly infested with 100 or more workers. The 1.0% permethrin impregnated containers of all sizes were very effective at excluding IFA through 16 months, and only allowed a few workers to get into the pots after that time. This only occurred in the 1 gallon containers until the 36 month evaluation of non-Windmill containers, possibly indicating that the 1 gallon untreated container was not of sufficient size to contain the entire colony. Data supplied by the 0.75% rate does not appear to be as effective at 21 and 24 months as the 1.0% rate.

Figure 1. Diagram of test arena.

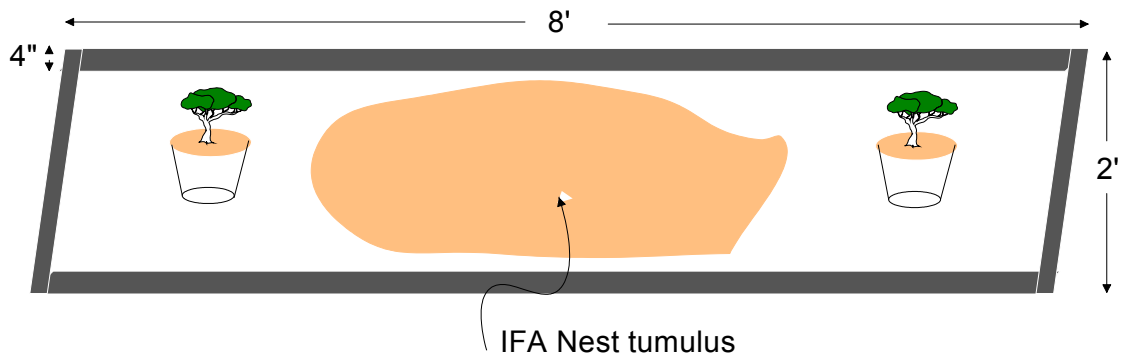


Figure 2. Percent of insecticide impregnated 1 gallon pots infested with IFA - Trial at Windmill Nursery - initiated 1999.

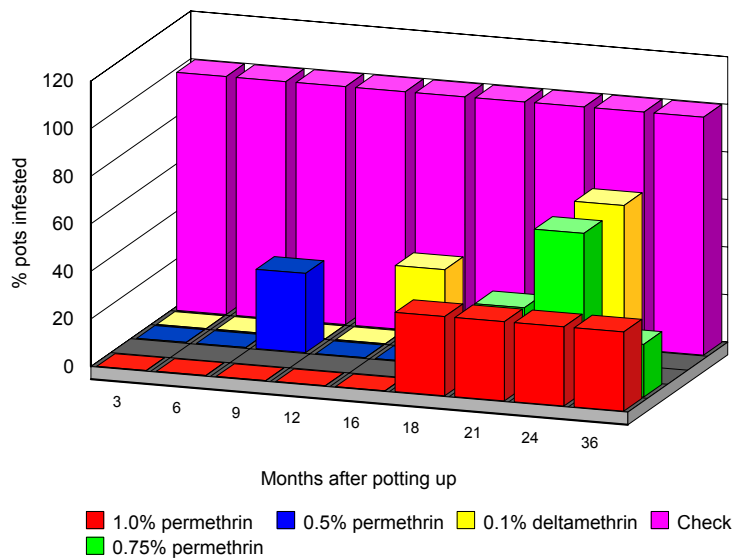


Figure 3. Percent of insecticide impregnated 3 gallon pots infested with IFA - Trial at Windmill Nursery - initiated 1999.

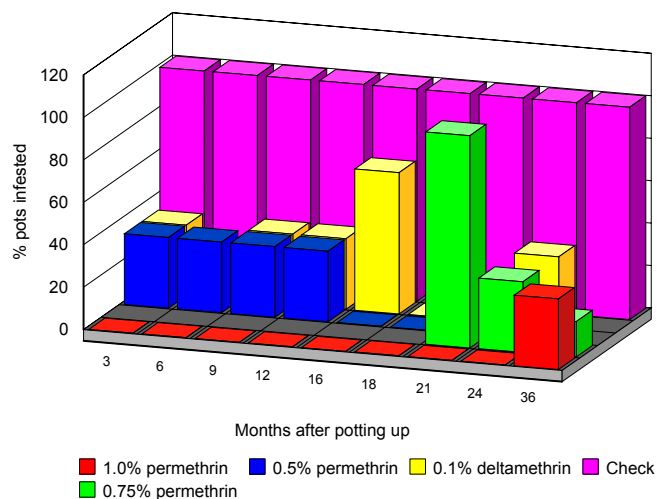
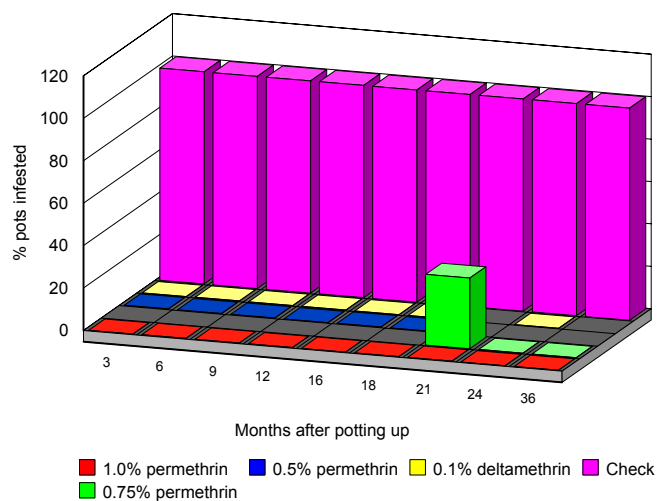


Figure 4. Percent of insecticide impregnated 10 gallon pots infested with IFA - Trial at Windmill Nursery - initiated 1999.



PROJECT NO: A1P04

PROJECT TITLE: Balled-and-Burlapped (B&B) Drench Treatments: Efficacy of Single Drench Treatments of Harvested Balls, 2001-2002.

REPORT TYPE: Interim

PROJECT LEADER/PARTICIPANT(s): Lee McAnally

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for balled and burlapped plants include immersion in a chlorpyrifos solution or twice daily drenches for three consecutive days with a chlorpyrifos solution. Adding additional chemical options to these use patterns will give growers more products to choose from. We also intend to investigate the efficacy of single drenches on balls instead of the labor intensive 3-day drenching schedule.

Initial trials were conducted to determine what pesticides might be good candidates for root ball drenches. Once pesticides were selected they were subjected to actual root ball drench trials.

MATERIALS AND METHODS:

Initial Container Trials:

Three gallon nursery containers were filled with sandy clay topsoil from southern Mississippi and placed on a brick in 12" x 18" x 5" plastic pan. The sides of the pans were painted with fluon to prevent escape. Three replicates per treatment were utilized. Field collected colonies were then separated from their nest tumulus by placing them in 2' x 8' x 5" separation trays and spreading the nest tumulus out. Artificial nests were provided for the colonies to move into. Once colonies moved into the artificial nests they were removed and placed into a 3 gallon bucket, anesthetized with CO₂ and 100cc of workers and brood were added to each soil filled pot. The ants were then allowed 24 hours to acclimate. Each pot was then treated with an amount of insecticidal solution equal to 1/5 the volume of the pot. Treatment rates were as follows:

Talstar F (bifenthrin)	0.1 lb ai/100 gal H ₂ O	45.2 ppm (1X)
Talstar F (bifenthrin)	0.05 lb ai/100 gal H ₂ O	22.6 ppm (½X)
Deltagard SC (deltamethrin)	0.08 lb ai/100 gal H ₂ O	35.5 ppm (1X)
Deltagard SC (deltamethrin)	0.04 lb ai/100 gal H ₂ O	17.75 ppm (½X)
Scimitar CS (lambda-cyhalothrin)	0.14 lb ai/100 gal H ₂ O	59.7 ppm (1X)
Scimitar CS (lambda-cyhalothrin)	0.07 lb ai/100 gal H ₂ O	29.85 ppm (½X)
Demon EC (cypermethrin)	0.07 lb ai/100 gal H ₂ O	50 ppm
Demon EC (cypermethrin)	0.14 lb ai/100 gal H ₂ O	100 ppm
Platinum (thiamethoxam)	0.07 lb ai/100 gal H ₂ O	50 ppm
Platinum (thiamethoxam)	0.14 lb ai/100 gal H ₂ O	100 ppm

Pots were checked daily for mortality for 7 days or until 100% mortality. Once 100% mortality was achieved the pots were then placed in a simulated can yard with overhead irrigation. At intervals of 1 week, 2 weeks, 1 month, 2 months, and 3 months one pot from each treatment was placed in the separation trays mentioned above, along with a check pot with untreated media (option test). A field collected colony was then placed in the tray. The pots were checked daily to determine which pot the colony moved into.

Root Ball Drenches:

Balled and burlapped plants 15 to 18 inches in diameter were placed in a 2' x 4' x 6" tray lined with black plastic sheeting (three root balls per treatment (one per tray). The upper walls of the trays were coated with fluon to prevent escape. Field collected colonies were then separated from their nest tumulus using the floatation method (Banks et al. 1981) and 100cc of ants and brood were placed on the root ball and allowed 24 hours to move into the root balls. Three replicates per treatment were used. The root balls were then drenched with 1 gallon of solution, corresponding to the lb ai/100 gal rate of the containerized test, by pouring the solution very slowly over the top of the root ball. Root balls were observed daily for mortality. Once 100% mortality was achieved root balls were moved outside to weather naturally. Soil core samples were taken monthly and subjected to alate queen bioassay (Appendix I).

RESULTS:

Initial Container Trials:

All treatments except the Platinum treatment provided 100% mortality within 24 hours. Platinum treatments did not appear to have any mortality and thus was excluded from the option test. Results of the option test are summarized in Table 1. All ants moved into the available control container in all choice tests through 2 months. At 3 months, ca. 100 workers did infest the high rate deltamethrin container, but all others continued to exclude infestation.

Root Ball Drenches:

To date, three trials have been initiated; Talstar $\frac{1}{2}X$, DeltaGard $\frac{1}{2}X$, and Scimitar 1X. All rates provided 100% mortality of colonies infesting the root ball. In alate female bioassays, the Talstar $\frac{1}{2}X$ drench rate produced 100% mortality through 4 months and 85% or better through 6 months. The Deltagard $\frac{1}{2}X$ drench rate produced 100% mortality through 6 months. The Scimitar 1X drench rate has produced 100% mortality through 5 months. Results of alate female bioassays are summarized in Table 2.

References Cited:

Banks, W.A., C.S. Lofgren, D.P. Jouvenaz, C.E. Stringer, P.M. Bishop, D.F. Williams, D.P. Wojcik and B.M. Glancey. 1981. Techniques for collecting, rearing, and handling imported fire ants. USDA, ARS, Science and Education Administration, Advances in Agricultural Technology, Southern Series, No. 21.

Table 1. Drench Option Test (number of ants moved into indicated pot after 7 days exposure*)

Treatment	Post-Treatment Interval									
	1 wk		2 wks		1 month		2 months		3 months	
	treated	check	treated	check	treated	check	treated	check	treated	check
Talstar 45.2 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Talstar 22.6 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Deltagard 35.5 ppm	0	5000	0	5000	0	5000	0	5000	100	4900
Deltagard 17.75 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Scimitar 59.7 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Scimitar 29.85 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Demon 50 ppm	0	5000	0	5000	0	5000	0	5000	0	5000
Demon 100 ppm	0	5000	0	5000	0	5000	0	5000	0	5000

*Assumes 5000 ants per tray

Table 2. Residual Activity of Various Drenches as Root ball Drenches

Treatment	% Mortality After 7 Days Exposure at Indicated Post-treatment Interval														
	1 Month					2 Months					3 months				
	Treated Replicate			AVG	Check	Treated Replicate			AVG	Check	Treated Replicate			AVG	Check
	1	2	3												
Talstar ½ label	100	100	100	100	0	100	100	100	100	0	100	100	100	100	0
Deltagard ½ label	100	100	100	100	0	100	100	100	100	0	100	100	100	100	0
Scimitar 1X label	100	100	100	100	0	100	100	100	100	0	100	100	100	100	0
Treatment	% Mortality After 7 Days Exposure at Indicated Post-treatment Interval														
	4 Months					5 Months					6 months				
	Treated Replicate			AVG	Check	Treated Replicate			AVG	Check	Treated Replicate			AVG	Check
	1	2	3												
Talstar ½ label	100	100	100	100	0	100	55	100	85	10	100	95	100	98.333	5
Deltagard ½ label	100	100	100	100	0	100	100	100	100	0	100	100	100	100	0
Scimitar 1X label	100	100	100	100	0	100	100	100	100	0					

PROJECT NO: A1P04

PROJECT TITLE: Alternative B&B Immersion or Drench Treatments for use in the IFA Quarantine, 2002

REPORT TYPE: Interim

PROJECT LEADER/PARTICIPANT(s): Lee McAnally, Shannon James; Jason Oliver and Nadeer Youssef of Tennessee State University

INTRODUCTION:

The Federal Imported Fire Ant Quarantine Program (7CFR §301.81) states that all regulated products (nursery stock) leaving the quarantined area must be treated in a prescribed manner. Currently, treatments for balled and burlaped (B&B) plants include immersion in a chlorpyrifos solution or twice daily drenches for three consecutive days with a chlorpyrifos solution. Several trials were initiated in conjunction with the Tennessee State University Nursery Crop Research Station to identify other dip or drench treatments that could be used concurrently for both the IFA Quarantine and Japanese Beetle programs.

MATERIALS AND METHODS:

Spring 2002 Dip Trial:

Due to limitations of resources, all treatments were conducted at the Japanese Beetle rates of application. Treatments were conducted in March 2002 at LanTenn Nursery (Belvidere, TN) by personnel from the Tennessee State University Nursery Crop Research Station. B&B plants were dipped in insecticidal solutions in large garbage cans lined with plastic. Treated plants were placed in a nursery environment. Core samples were taken and shipped to the CPHST-ANPCL soil Inhabiting Pests Lab where they were subjected to standard alate queen bioassay (Appendix I). Core samples were taken at 60, 120 and 180 days post-treatment. Candidate treatments and rates were as follows:

Treatment	Common Name	Rate (lb ai/100 gal H ₂ O)
Mach 2 2SC	Benzoic acid	1.50 (1X)
		0.75 (1/2X)
Orthene 75WSP	Acephate	.075 (1X)
Talstar Nursery F	Bifenthrin	0.23 (1X)
		0.12 (1/2X)
Marathon 60WP	Imidachloprid	0.30 (1X)
		0.15 (1/2X)
Dylox 80 T&O	Dimethyl phosphonate	8.00 (1X)
		4.00 (1/2X)
Sevin SL	Carbaryl	8.00 (1X)
		4.00 (1/2X)
Flagship 25WG	Thiamethoxam	0.13 (1X)

		0.07 (1/2X)
Deltagard GC 5S	Deltamethrin	0.13 (1X)
Dursban TNP	Chlorpyrifos	2.00 (1X)
Control		-----

Fall 2002 Dip and Drench Trials:

Again, rates of application were based on Japanese beetle rates, due to the dual nature of this testing. Fall dip trials were conducted in the same manner and location as above in late October 2002. Additionally, B&B Dogwood trees were drenched in the field twice a day for 3 consecutive days. Drench solutions were mixed in approx. 35 gal drums lined with polyethylene drum liners. Solutions were applied by means of a hand operated transfer pump fitted with a garden hose and a spray wand with a showerhead type spray nozzle. Approximately 1/6 gal of solution was applied to each root ball at each application time for a total of 1 gallon of solution per ball. At 2 weeks, and then at monthly intervals core samples were taken from four replicates of each treatment and shipped to the Gulfport Plant Protection Station where they were subjected to standard alate queen bioassay. Core samples from the dipped plants were taken from the top of the root ball. Core samples from the drenched plants were taken from the top, middle and bottom of the root ball as it was lying in the field.

Treatment	Common name	Rate (lb ai/100 gal H ₂ O)
Talstar Nursery F	Bifenthrin	0.23 (1X)
		0.12 (1/2X)
Flagship 25WG	Thiamethoxam	0.13 (1X)
		0.07 (1/2X)
Deltagard GC 5S	Deltamethrin	0.13 (1X)
Marathon 60WP	Imidachloprid	0.40 (1X)
Dursban TNP	Chlorpyrifos	2.00 (1X)
Scimitar SC	Lambda-cyhalothrin	0.034 (1X)
Control		-----

RESULTS:

Spring 2002 Dip Trial:

Both full and half rates of Talstar F were 100% effective against IFA alate females for 6 months after treatment (Figure 1). Dursban and the full rate of Flagship were very effective for 4 months. DeltaGard and the half rate of Flagship were effective for 2 months.

Fall 2002 Dip and Drench Trials:

Dip: Only cores from the top of the ball were evaluated for dip treatments, assuming that because the balls were completely immersed in the treatment solution all parts of the ball should be equally treated. At 3 months after treatment, all treatments and rates have been 100% effective. Through 2 months all rates provided 100% control within 6 days of exposure, while at 3 months, both Flagship rates required up to 14 days to provide 100% control (still acceptable).

Drench: In this trial, the top of the root ball was considered that area which received the watering in or drench treatment, and the bottom was that area opposite the “top”, usually the side

in contact with the ground. All products and rates from media collected from the top of the root balls provided excellent control of IFA alate females through 3 months (Figure 3). Media collected from the middle of the balls was very ineffective at 2 weeks and was not collected after that time. However, in hindsight, this should have been done since many of these products appeared to penetrate the media over time (as shown in the bottom data) and provide better control later in the evaluation period. Media collected from the bottom of drenched root balls provided interesting and erratic information for many of the products (Figure 5). However, the high rate of Talstar consistently provided >90% control of alate females through 3 months. The Dursban was less effective at 2 weeks than at any other time, and provided 100% control at 1 month, dropping to around 90% at months 2-3, verifying current treatment certification period of 30 days.

Testing will continue for a few more months. The most promising treatments will be replicated in spring 2003 in Tennessee and additional trials in Mississippi, using IFA rates were applicable, will also be conducted in 2003.

Figure 1. Efficacy of various immersion/dip treatments for B&B nursery stock – spring 2002.

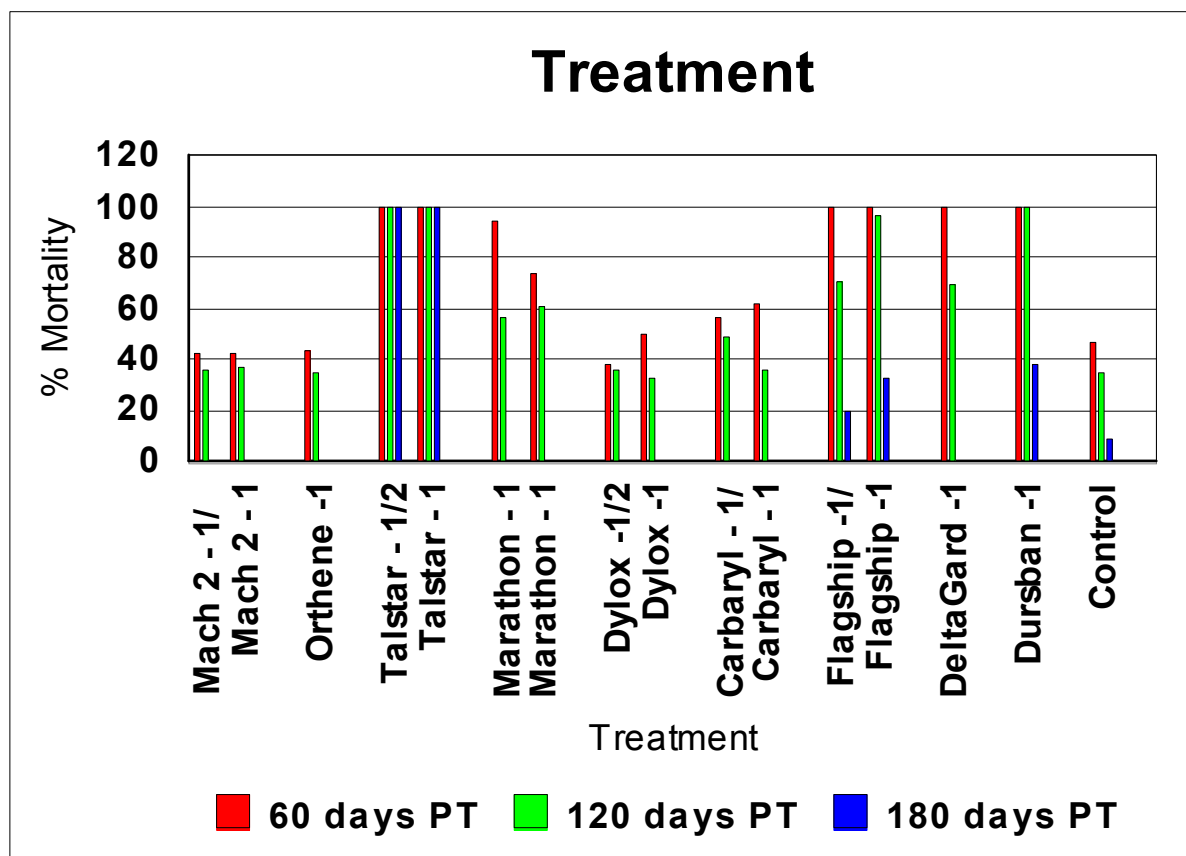


Figure 2. Efficacy of various immersion/dip treatments for B&B nursery stock – fall 2002.

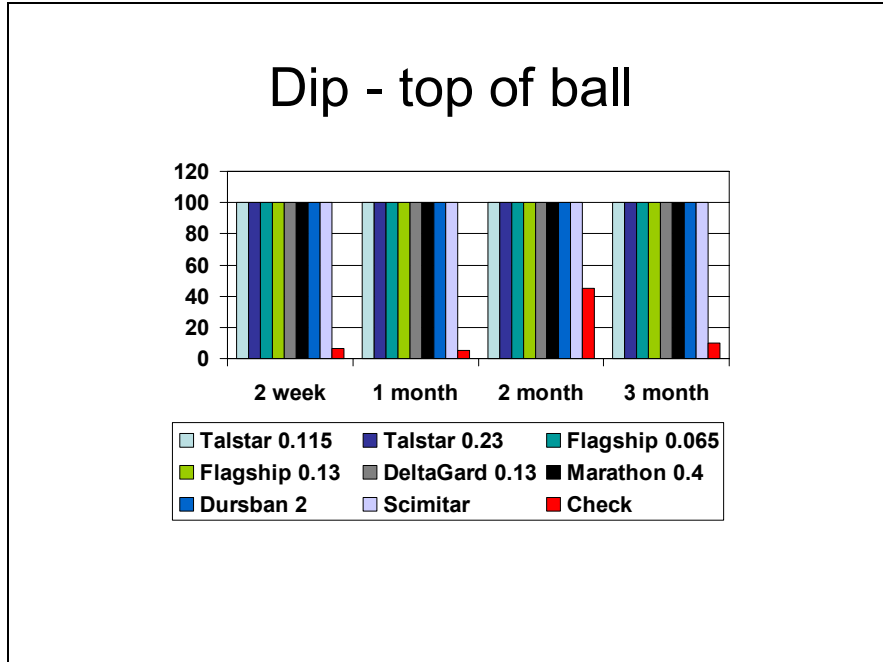


Figure 3. Efficacy of B&B drenches (2X/day on 3 consecutive days) testing media collected from various parts of the root ball – top of ball.

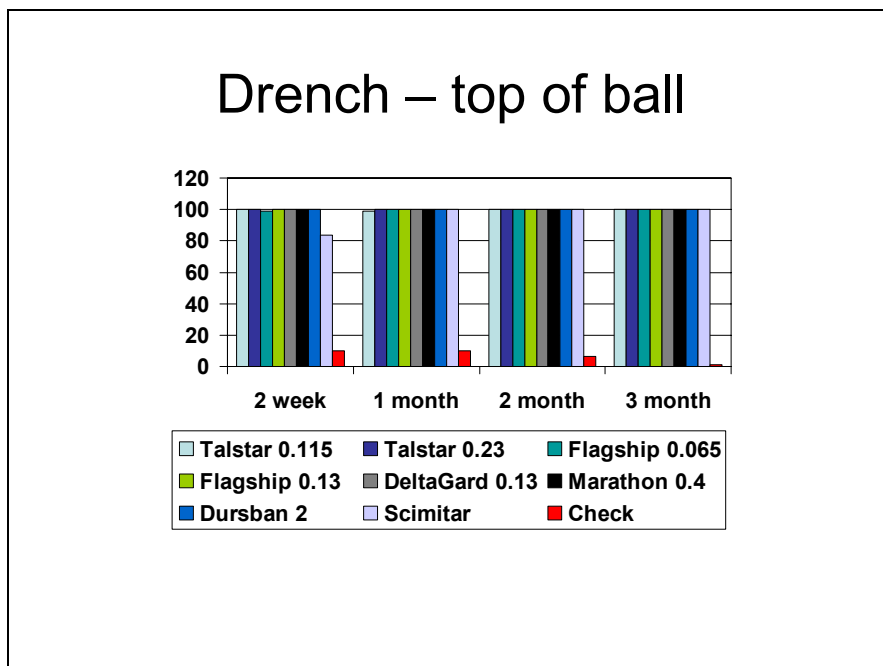


Figure 4. Efficacy of B&B drenches (2X/day on 3 consecutive days) testing media collected from various parts of the root ball – middle of ball.

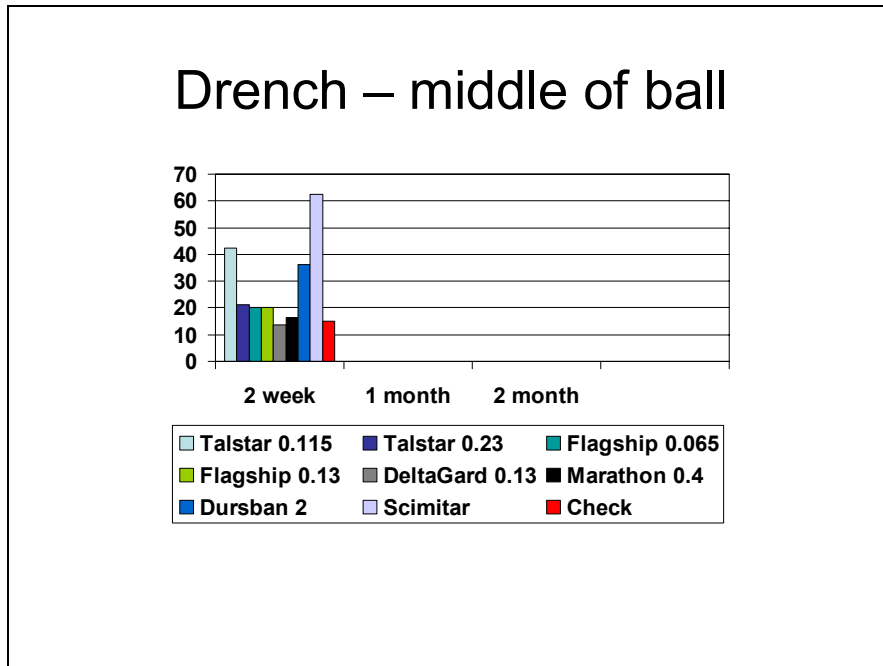
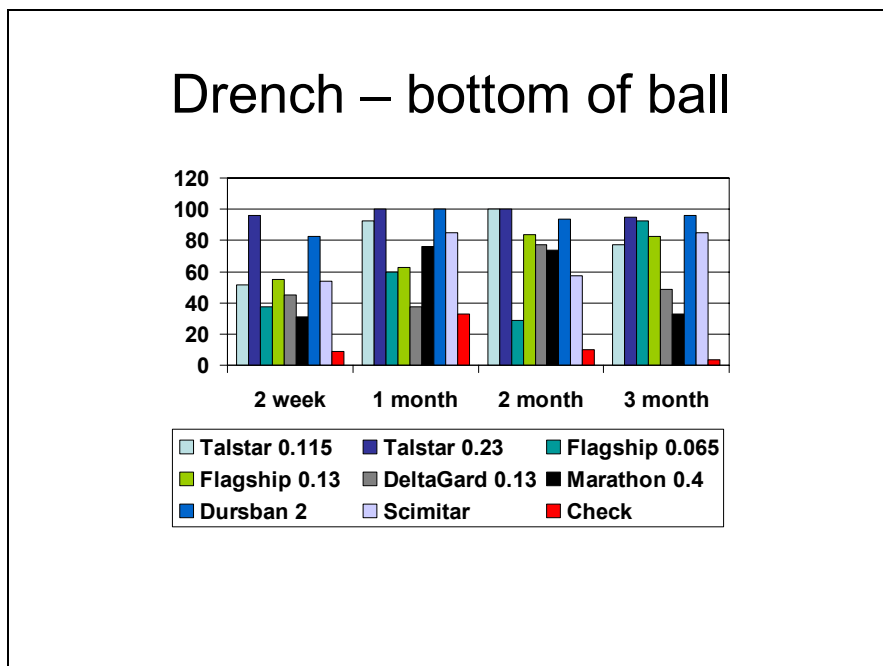


Figure 5. Efficacy of B&B drenches (2X/day on 3 consecutive days) testing media collected from various parts of the root ball – bottom of ball.



PROJECT NO: A1P04 - GPPS02-01

PROJECT TITLE: Development of Alternative Quarantine Treatment for Field Grown Nursery Stock – Surface Band Application, 2001

TYPE REPORT: Final

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade

INTRODUCTION:

APHIS is responsible for developing treatment methodologies for certification of regulated items, such as field grown/balled-and-burlapped (B&B) nursery stock in the Imported Fire Ant Quarantine. Current treatments for field grown stock, as described below, are labor intensive, cumbersome and expensive. The only in-field treatment utilizes chlorpyrifos, the future of which is uncertain at best. Thus additional treatment methods are needed to insure movement of this type of material. Imported fire ants are slowly moving into areas of Tennessee where many producers of field grown nursery stock are located. This has prompted renewed interest in development of new treatments for this stock. New regulatory treatment methods for field grown/B&B nursery stock are needed to insure that nursery growers can compile with the Federal IFA Quarantine.

Currently the Federal Imported Fire Ant Quarantine (7CFR 301.81) has three treatment regimens for certification of field-grown and B&B nursery stock. The in-field treatment requires a broadcast application of an approved bait followed in 3-5 days by a broadcast application of granular chlorpyrifos. After a 30-day exposure period, plants are certified for 12 weeks. In 1999, PPQ allowed for a second application of the granular chlorpyrifos to extend the certification period for an additional 12 weeks. For harvested B&B stock, there are two certification methods: immersion in a chlorpyrifos solution or watering twice daily with a chlorpyrifos solution for 3 consecutive days.

The in-field treatment currently requires that the treatment must extend at least 10 feet from the base of each plant to be certified. This virtually means that the treatments must be applied broadcast to the entire nursery block. Numerous granular formulations of common insecticides such as diazinon, chlorpyrifos, acephate, and others are labeled for spot treatment of imported fire colonies. Imported fire ant colonies readily respond to any insecticide application made directly to the nest by relocating the colony (Collins & Callcott 1995, Hays et al. 1982, Franke 1983, Williams & Lofgren 1983). This insecticide induced movement is usually over a relatively short distance (1.5 to 3.0 meters), but can be greater (AMC, personal observation). The primary objective of a quarantine treatment for field grown nursery stock is to render the plants fire ant free. Therefore, it does not matter if colonies are killed outright by the treatment or simply induced to move away from the area around each individual plant intended for harvest.

MATERIALS AND METHODS:

Preliminary testing was initiated in Sept. 2001 by testing several liquid and granular insecticides against individual IFA mounds in the field. Treatments were applied to 10 mounds each in a pasture in Harrison Co., Mississippi on Sept. 7, 2001. Evaluations were made at 3 days after treatment, and weekly thereafter. Results of this trial indicated which insecticides should move on to the next phase of the project (see GPPS01-02 report).

Mississippi fall 2001 preliminary trial: The second phase of this project was to evaluate promising insecticides as band treatments for use in in-field nursery stock situations. Growers routinely use band treatments for fertilizer and herbicide treatments. Treatments were applied at Camp Shelby, Mississippi on October 22, 2001. Granular band treatments were applied using a Gandy granular band spreader attached to a farm tractor. Liquid band treatments were applied using a roller pump boom sprayer equipped with the appropriate number of TKSS tips to provide various swath widths, and a total spray volume equivalent to ca. 38 gal/acre. Band widths for the preliminary trial were 30" to 36" bands on either side of the "stock" for a treated width of 5' to 6'. Treatments were applied to plots 5' to 6' wide (depending on swath width tested) and approx. 1000' long (long enough to include a minimum of 6-8 IFA mounds per plot). There were 3 replicates per treatment, including an untreated check, unless noted. Prior to treatment and at 1, 2, and 4 weeks after final treatment, active IFA colonies in each plot were enumerated. Treatments were evaluated at 4-week intervals thereafter.

<u>Chemical Formulation</u>	<u>Rate of Application</u>
Talstar G (0.2%)	200 lb/acre (0.4 lb ai/acre)
Fipronil G (0.0143%)	87 lb/acre (0.0125 lb ai/acre)
DeltaGard G (0.1%)	131 lb/acre (0.13 lb ai/acre)
Talstar F (7.9%)	40 oz/acre (0.2 lb ai/acre)
DeltaGard SC (4.75%)	39 oz/acre (0.13 lb ai/acre)

Insecticides with promising results in the preliminary fall 2001 Mississippi trial will be repeated in Mississippi and Tennessee in 2002-2003 and additional chemicals added to the project as warranted.

RESULTS:

Mississippi fall 2001 preliminary trial: There was no rainfall in the 1-2 weeks preceding the application of this trial, and no rain fell in the first 3 weeks after treatment. The first rainfall, of 3.5", occurred just prior to the 4-week evaluation. Thereafter, rainfall occurred weekly throughout the trial.

There were no significant decreases in colony numbers until after the first rainfall, indicating that all treatments, including the liquid sprays, were more effective after "watering in" (Figure1). In future trials, a greater volume of finished spray will be applied to enhance the insecticide activity. As in individual mound trials, the fipronil granular was very slow to eliminate or repel IFA from the treatment area. All other treatments reduced the number of active IFA mounds within the treatment area by ca. 90% 4-6 weeks after treatment. The granular formulations were

more consistent in providing continuous control in the treatment area of >90% for ca. 10 weeks after achieving control. However, all were somewhat erratic and not of quarantine efficacy.

Refinement of application techniques and additional testing is required for this treatment alternative.

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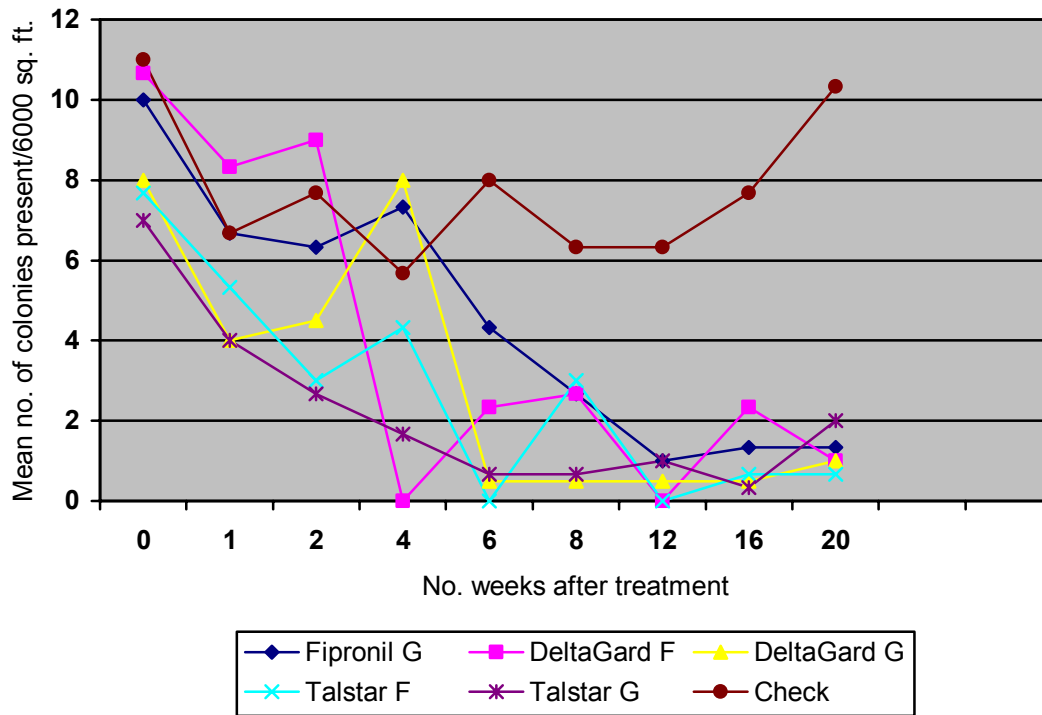
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Hays, S. B., P. M. Horton, J. A. Bass and D. Stanley. 1982. Colony movement of imported fire ants. *J. Georgia Entomol. Soc.* 17: 266-272.

Williams, D. F. and C. S. Lofgren. 1983. Imported fire ant control: evaluation of several chemicals for individual mound treatments. *J. Econ. Entomol.* 76: 1201-1205.

Figure 1. Activity of various liquid and granular insecticides applied in a band treatment against imported fire ants.



PROJECT NO: A1P04 - GPPS02-02

PROJECT TITLE: Development of Alternative Quarantine Treatment for Field Grown Nursery Stock – Surface Band Application, 2002

TYPE REPORT: Final

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade and Craig Hinton (NMRAL aide)

INTRODUCTION:

APHIS is responsible for developing treatment methodologies for certification of regulated items, such as field grown/balled-and-burlapped (B&B) nursery stock in the Imported Fire Ant Quarantine. Current treatments for field grown stock, as described below, are labor intensive, cumbersome and expensive. The only in-field treatment utilizes chlorpyrifos, the future of which is uncertain at best. Thus additional treatment methods are needed to insure movement of this type of material. Imported fire ants are slowly moving into areas of Tennessee where many producers of field grown nursery stock are located. This has prompted renewed interest in development of new treatments for this stock. New regulatory treatment methods for field grown/B&B nursery stock are needed to insure that nursery growers can compile with the Federal IFA Quarantine.

Currently the Federal Imported Fire Ant Quarantine (7CFR 301.81) has three treatment regimens for certification of field-grown and B&B nursery stock. The in-field treatment requires a broadcast application of an approved bait followed in 3-5 days by a broadcast application of granular chlorpyrifos. After a 30-day exposure period, plants are certified for 12 weeks. In 1999, PPQ allowed for a second application of the granular chlorpyrifos to extend the certification period for an additional 12 weeks. For harvested B&B stock, there are two certification methods: immersion in a chlorpyrifos solution or watering twice daily with a chlorpyrifos solution for 3 consecutive days.

The in-field treatment currently requires that the treatment must extend at least 10 feet from the base of each plant to be certified. This virtually means that the treatments must be applied broadcast to the entire nursery block. Numerous granular formulations of common insecticides such as diazinon, chlorpyrifos, acephate, and others are labeled for spot treatment of imported fire colonies. Imported fire ant colonies readily respond to any insecticide application made directly to the nest by relocating the colony (Collins & Callcott 1995, Hays et al. 1982, Franke 1983, Williams & Lofgren 1983). This insecticide induced movement is usually over a relatively short distance (1.5 to 3.0 meters), but can be greater (AMC, personal observation). The primary objective of a quarantine treatment for field grown nursery stock is to render the plants fire ant free. Therefore, it does not matter if colonies are killed outright by the treatment or simply induced to move away from the area around each individual plant intended for harvest.

MATERIALS AND METHODS:

Preliminary testing was initiated in Sept. 2001 by testing several liquid and granular insecticides against individual IFA mounds in the field. Results of this trial indicated promising results with acephate, Talstar, and DeltaGard (see GPPS01-02 report). A second preliminary trial, taking selected products to the field and testing in band applications was initiated in fall 2001 in Mississippi. Using band widths of 5' to 6', Talstar and DeltaGard, both liquid and granular formulations showed promising results (see GPPS02-01).

Treatments were applied at Hattiesburg Municipal Airport, Mississippi on April 29-30, 2002. Granular band treatments were applied using a Gandy 48" granular drop spreader attached to a farm tractor. Liquid band treatments were applied using a roller pump boom sprayer equipped with two standard flat spray tips (8015-SS; TeeJet Corp.) to provide various a 36" band spray and a total spray volume equivalent to ca. 76 gal/acre. Band widths for the preliminary trial were 36" bands on either side of the "stock" for a treated width of 6' for the liquid applications and 48" bands on either side of the "stock" for a treated width of 8' for the granular treatments. Treatments were applied to plots x feet wide (depending on swath width tested) and 800' long (long enough to include a minimum of 5 IFA active mounds per plot). Active mounds within two-feet of the center line of the treatment were counted, leaving a treated buffer area outside the area that would normally be harvested with B&B stock. There were 3 replicates per treatment, including an untreated check. Prior to treatment and at 1, 2, 4, 6 and 8 weeks after final treatment, active IFA colonies in each plot were enumerated. Treatments were evaluated at 4-week intervals thereafter. Mounds were evaluated using as little disturbance as possible, usually a wire flag inserted into the mound. Mounds were considered active if any workers appeared. Using this data, colony mortality was calculated and experimental data was statistically analyzed using analysis of variance, and treatment means separated using the LSD test ($P=0.05$) for each posttreatment rating interval (see GPPS02-01 report).

<u>Chemical Formulation</u>	<u>Rate of Application</u>
Talstar G (0.2%)	200 lb/acre (0.4 lb ai/acre)
DeltaGard G (0.1%)	131 lb/acre (0.13 lb ai/acre)
Talstar F (7.9%)	40 oz/acre (0.2 lb ai/acre)
DeltaGard SC (4.75%)	39 oz/acre (0.13 lb ai/acre)
Scimitar CS (9.7%)	10 oz/acre (0.06875 lb ai/acre)
Sevin SL (43.0%)	39 oz/acre (1.22 lb ai/acre)

RESULTS:

The Sevin was inadvertently applied at the same bulk rate as the DeltaGard SC instead of the labeled rate of 130.68 oz/acre (ca. 4 lb ai/acre).

In the first 4 weeks of the trial there was only 1.6 inches of rainfall, with no rainfall the 2 weeks prior to treatment (Figure 1). Rainfall in the figure shown on a specific date is actually the amount of rainfall occurring during the previous 2-4 weeks (depending on the evaluation interval). Temperatures during the first 4 weeks were above normal. There was a good rainfall

between the 4 and 6 week evaluations (3.4 inches). The unusually hot and dry April and May severely affected the control plots in this trial. The evaluation method used here, minimal disturbance, does not allow us to dig down into the mound during hot, dry periods to find workers that may be present, but deep within the mound. As shown by the data for weeks 2-4, very few mounds in the check plots were rated as active (Table 1 and Figure 1). Therefore, it is uncertain whether the insecticides had eliminated colonies in the treated areas or whether the ants were not detectable due to weather conditions. However, by 6 weeks after treatment, the check plots had somewhat recovered, and at 6, 8 and 12 weeks after treatment, all treatments were significantly different from the controls. The Talstar products were the most effective for the longest period of time, however all products were promising.

This trial along with previous work indicates that single band contact insecticide treatments are not sufficient as stand alone treatments for quarantine level efficacy. Future trials will incorporate a broadcast bait application to the entire area prior to the band treatments.

References Cited:

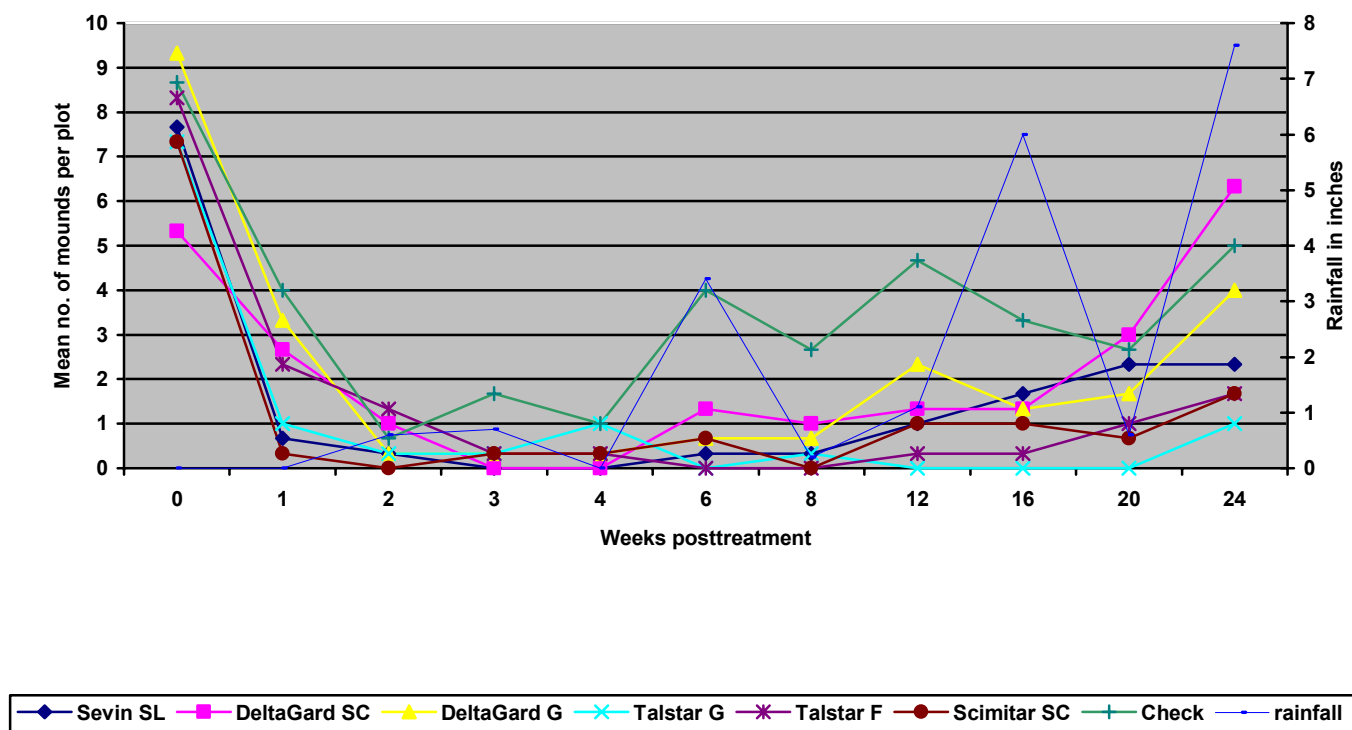
- Collins, H.L. and A-M. Callcott. 1995. Effectiveness of spot insecticide treatments on red imported fire ant control. *J. Entomol. Sci.* 30: 489-496.
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- Hays, S.B., P.M. Horton, J.A. Bass and D. Stanley. 1982. Colony movement of imported fire ants. *J. Georgia Entomol. Soc.* 17: 266-272.
- Williams, D.F. and C.S. Lofgren. 1983. Imported fire ant control: evaluation of several chemicals for individual mound treatments. *J. Econ. Entomol.* 76: 1201-1205.

Table 1. Efficacy of band treatments in eliminating IFA colonies from a specific area.

Treatment	Mean no. mounds pretreat*	Mean % change in no. active mounds present from pretreatment nos. at indicated weeks posttreatment									
		1	2	3	4	6	8	12	16	20	24
DeltaGard G	9.33	-58.61ab	-97.78ab	-95.38a	-95.83a	-91.11ab	-93.61ab	-78.06b	-86.67ab	-82.50ac	-56.11a
DeltaGard SC	5.33	-48.89b	-82.22ab	-100.0a	--	-75.56bc	-81.11b	-74.44b	-74.44ab	-43.33b	+22.22b
Scimitar SC	7.33	-95.24a	-100.0a	-95.24a	-95.24a	-90.48ab	-100.0a	-88.57ab	-87.14ab	-91.90a	-81.90a
Sevin SL	7.67	-92.59a	-95.83ab	-100.0a	-100.0a	-95.83ab	-95.83a	-86.57ab	-79.17ab	-68.06c	-68.52a
Talstar F	8.33	-63.01ab	-76.67b	-93.33ab	-93.33a	-100.0a	-100.0a	-97.62a	-97.62a	-88.57ac	-81.75a
Talstar G	7.33	-85.71ab	-95.24ab	-95.24a	-88.57a	-100.0a	-96.67a	-100.0a	-100.0a	-100.0a	-87.14a
Check	8.67	-59.09ab	-92.59ab	-81.65b	-89.56a	-57.74c	-66.13c	-45.96c	-61.11b	-68.52c	-41.07a

Means within a column followed by the same letter are not significantly different (LSD, $P < 0.05$)

Figure 1. Colony mortality after a broadcast treatment of bait followed by a band treatment of contact insecticide.



PROJECT NO: A1P04

PROJECT TITLE: Development of Alternative Quarantine Treatment for Field Grown Nursery Stock – Broadcast Bait plus Surface Band Application, Fall 2002

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade, Shannon James, Ron Weeks

INTRODUCTION:

APHIS is responsible for developing treatment methodologies for certification of regulated items, such as field grown/balled-and-burlapped (B&B) nursery stock in the Imported Fire Ant Quarantine. Current treatments for field grown stock, as described below, are labor intensive, cumbersome and expensive. The only in-field treatment utilizes chlorpyrifos, the future of which is uncertain at best. Thus additional treatment methods are needed to insure movement of this type of material. Imported fire ants are slowly moving into areas of Tennessee where many producers of field grown nursery stock are located. This has prompted renewed interest in development of new treatments for this stock. New regulatory treatment methods for field grown/B&B nursery stock are needed to insure that nursery growers can compile with the Federal IFA Quarantine.

Currently the Federal Imported Fire Ant Quarantine (7CFR 301.81) has three treatment regimens for certification of field-grown and B&B nursery stock. The in-field treatment requires a broadcast application of an approved bait followed in 3-5 days by a broadcast application of granular chlorpyrifos. After a 30-day exposure period, plants are certified for 12 weeks. In 1999, PPQ allowed for a second application of the granular chlorpyrifos to extend the certification period for an additional 12 weeks. For harvested B&B stock, there are two certification methods: immersion in a chlorpyrifos solution or watering twice daily with a chlorpyrifos solution for 3 consecutive days.

The in-field treatment currently requires that the treatment must extend at least 10 feet from the base of each plant to be certified. This virtually means that the treatments must be applied broadcast to the entire nursery block. Numerous granular formulations of common insecticides such as diazinon, chlorpyrifos, acephate, and others are labeled for spot treatment of imported fire colonies. Imported fire ant colonies readily respond to any insecticide application made directly to the nest by relocating the colony (Collins & Callcott 1995, Hays et al. 1982, Franke 1983, Williams & Lofgren 1983). This insecticide induced movement is usually over a relatively short distance (1.5 to 3.0 meters), but can be greater (AMC, personal observation). The primary objective of a quarantine treatment for field grown nursery stock is to render the plants fire ant free. Therefore, it does not matter if colonies are killed outright by the treatment or simply induced to move away from the area around each individual plant intended for harvest.

Preliminary testing was initiated in Sept. 2001 by testing several liquid and granular insecticides against individual IFA mounds in the field. Results of this trial indicated promising results with acephate, Talstar, and DeltaGard (see GPPS01-02). A second preliminary trial, taking selected products to the field and testing in band applications was initiated in fall 2001 and spring 2002 in Mississippi. Using band widths of 5' to 6', Talstar and DeltaGard, both liquid and granular formulations showed promising results (see GPPS02-01; GPPS02-02). These trials, while promising, showed that contact insecticide treatments alone were not effective enough for use in the IFA quarantine. Therefore, the following trial was initiated applying a broadcast bait application followed by the contact insecticide applied as a band treatment.

MATERIALS AND METHODS:

Bait treatments were applied at Hattiesburg Municipal Airport, Mississippi on October 31, 2002 using a shop built spreader on a farm tractor calibrated to apply 1.25 lbs/acre. Due to excessive rainfall over the following weeks, the contact insecticides were not applied until November 25, 2002. Granular band treatments were applied using a Gandy 48" granular drop spreader attached to a farm tractor. Liquid band treatments were applied using a roller pump boom sprayer equipped with two standard flat spray tips (8015-SS; TeeJet Corp.) to provide various a 36" band spray and a total spray volume equivalent to ca. 76 gal/acre. Band widths for the preliminary trial were 36" bands on either side of the "stock" for a treated width of 6' for the liquid applications and 48" bands on either side of the "stock" for a treated width of 8' for the granular treatments. Treatments were applied to plots x feet wide (depending on swath width tested) and 800' long (long enough to include a minimum of 5 IFA active mounds per plot). Active mounds within two-feet of the center line of the treatment were counted, leaving a treated buffer area outside the area that would normally be harvested with B&B stock. There were 3 replicates per treatment, including an untreated check. Prior to treatment and at 1, 2, 4, 6 and 8 weeks after final treatment, active IFA colonies in each plot were enumerated. Treatments were evaluated at 4-week intervals thereafter. Mounds were evaluated using as little disturbance as possible, usually a wire flag inserted into the mound. Mounds were considered active if any workers appeared. Using this data, colony mortality was calculated and experimental data was statistically analyzed using analysis of variance, and treatment means separated using the LSD test ($P=0.05$) for each posttreatment rating interval. Original plans were to include more contact insecticides than the four listed below, but excessive rainfall flooded areas of the test site and treatments could not be made.

<u>Chemical Formulation</u>	<u>Rate of Application</u>
Talstar G (0.2%)	200 lb/acre (0.4 lb ai/acre)
DeltaGard G (0.1%)	131 lb/acre (0.13 lb ai/acre)
Talstar F (7.9%)	40 oz/acre (0.2 lb ai/acre)
DeltaGard SC (4.75%)	39 oz/acre (0.13 lb ai/acre)

RESULTS:

Both granular formulations provided 100% control of IFA in the evaluation area within 2 weeks of the contact insecticide application and maintained that control through 15 weeks (Table 1 and Figure 1). DeltaGard SC reached and maintained 100% control at 5 weeks and through 15

weeks. The Talstar F reached and maintained 95% control the first week and maintained that control through 15 weeks. One colony in one Talstar F plot remained active throughout the test period, however it has become significantly less active over time, with very few workers appearing at the 15 week evaluation. This colony was located next to (against) the small concrete structure supporting a wind sock and apparently our application did not completely saturate this area, and certainly did not overlap with the “other side” of the application. This lone remaining colony, although weakened stresses the importance of complete and accurate application of the contact insecticide in this “dual” treatment technique. Evaluations will continue until reinfestation occurs and additional trials will be initiated to confirm efficacy of this type of treatment for infield quarantine treatment.

References Cited:

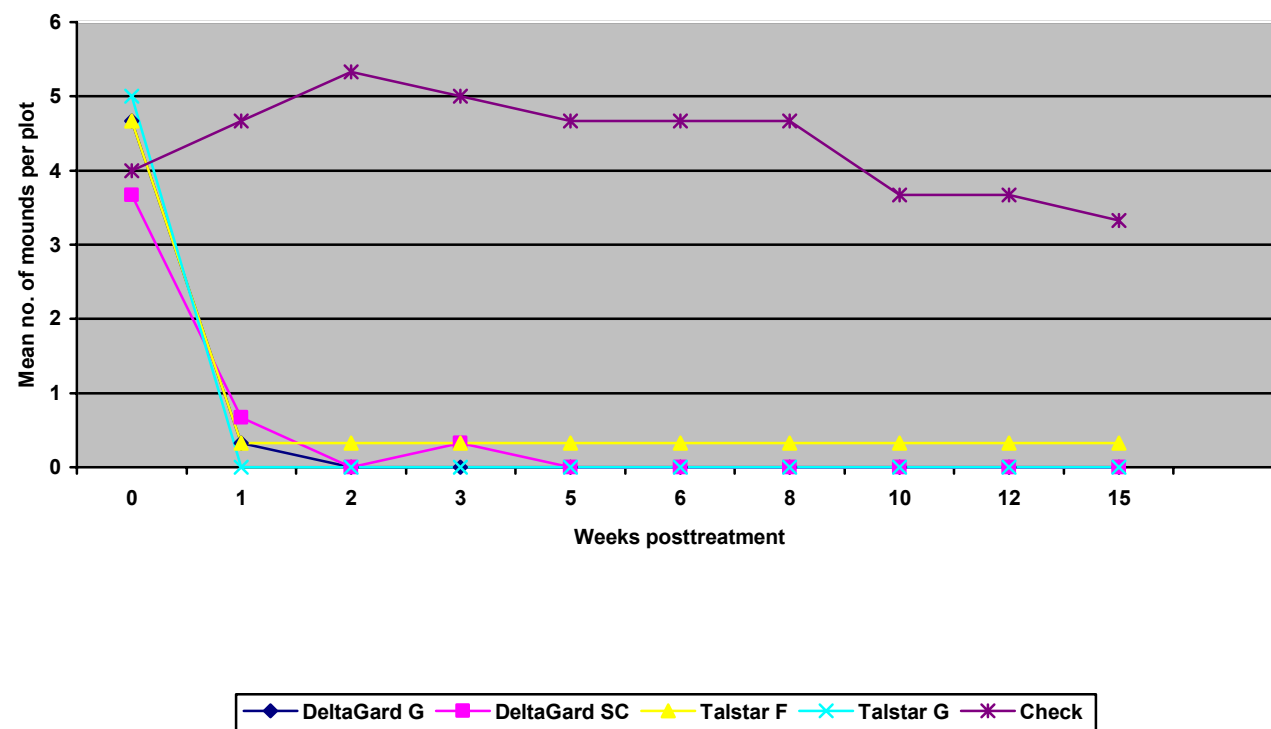
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- Franke, O.F. 1983. Efficacy of tests of single mound treatments for control of red imported fire ants. *Southwest. Entomol.* 8: 42-45.
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- Williams, D.F. and C.S. Lofgren. 1983. Imported fire ant control: evaluation of several chemicals for individual mound treatments. *J. Econ. Entomol.* 76: 1201-1205.

Table 1. Efficacy of band treatments in eliminating IFA colonies from a specific area.

Treatment	Mean no. mounds pretreat*	Mean % change in no. active mounds present from pretreatment nos. at indicated weeks posttreatment										
		1	2	3	5	6	8	10	12	15		
DeltaGard G	4.67a	-94.4a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a		
DeltaGard SC	3.67a	-83.3a	-100.0a	-91.7a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a		
Talstar F	4.67a	-95.2a	-95.2a	-95.2a	-95.2a	-95.2a	-95.2a	-95.2a	-95.2a	-95.2a		
Talstar G	5.00a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a		
Check	4.00a	36.1b	58.3b	41.7b	25.0b	33.3b	25.0b	-2.8b	-2.8b	-11.1b		

Means within a column followed by the same letter are not significantly different (LSD, $P < 0.05$)

Figure 1. Colony mortality after a broadcast treatment of bait followed by a band treatment of contact insecticide.



PROJECT NO: A1P04 - GPPS01-01

PROJECT TITLE: Traditional In-field Treatments for Field Grown Nursery Stock: Bait plus Broadcast Contact Insecticide, 2001

TYPE REPORT: Final

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade, Chris Doxey

INTRODUCTION:

Current quarantine treatments for field grown nursery include a bait treatment followed in 3-5 days by a granular chlorpyrifos treatment. This treatment allows for 12 weeks of certification after a 30-day exposure period. Treatment must extend 10 ft. from the base of each plant, ultimately meaning that entire blocks of stock must be treated. With the impending loss of chlorpyrifos, we initiated a trial to replace the chlorpyrifos portion of this treatment regimen with more available chemicals.

MATERIALS AND METHODS:

The bait used was Distance (Valent Corp., Walnut Creek, CA). The contact insecticides were Talstar 0.2G, Talstar Flowable, and fipronil 0.0143G. We also tested a blend of fipronil bait (1.5 ppm) and fipronil granule (0.0143%). Rates of application are shown below.

<u>Insecticide</u>	<u>Formulation</u>	<u>Rate of Application</u>
Distance	bait	1.5 lb/acre
Talstar	0.2G	200 lb/acre (0.4 lb ai/acre)
Talstar	Flowable	40 oz/acre (0.2 lb ai/acre)
fipronil	0.0143G	87 lb/acre (0.0125 lb ai/acre)
fipronil blend	15 lb bait + 87 lb granule	100 lb/acre

The test site was located at the Laurel Municipal Airport in Laurel, MS. As with many airport sites, the upkeep and accessibility is superior, but ant populations tend to be somewhat low. The bait was applied on June 18, 2001 with a shop built applicator mounted on a farm tractor. Air temperature was 85-87°F and the soil temperature was 72°F. Due to weather delays the contact insecticides were applied on June 21, 2001 and June 26, 2001. Air temperature was 87°F and soil temperature was 80°F on June 21 and 90°F and 80°F, respectively, on June 26. Granular material was applied with a Herd™ spreader mounted on a farm tractor. Liquid material was applied with a roller pump boom sprayer equipped with five TKSS tips with provided a 10 ft. swath. The system was operated at 50 psi providing 38 gallons of finished spray per acre. There were three replicates per treatment, and all test plots were 1.0 acre in size. A ¼-acre circular efficacy plot was established in the center of each 1.0 acre test plot. Prior to bait application and at 1, 2, and 4 weeks after final treatment (June 26), IFA populations in each efficacy plot were evaluated using the population index system developed by Harlan et al. (1981), and later revised

by Lofgren and Williams (1982). Treatments were evaluated at 4 week intervals thereafter. Using this data, both colony mortality and decrease in pretreatment population indices were calculated. Experimental data were statistically analyzed using analysis of variance, and treatment means were separated using the LSD test ($P=0.05$) for each posttreatment rating interval.

RESULTS:

Due to the difference in application times, evaluations were started 1 week after the last application. All treatments provided >90% mortality within 2 weeks of application of the granular/liquid insecticide, and by 4-5 weeks after treatment all treatments had provided 100% mortality (Tables 1 & 2). All treatments provided 100% control through 8-9 weeks after treatment and greater than 91% control through 44 weeks. High mortality in the check plots at weeks 12-17 is not unusual in south Mississippi in late summer. However, the continued high check mortality from late Nov through early May, when we expect to see an increase in ant populations, is unexplained.

RECOMMENDATION:

All treatments appeared to be excellent replacements for dursban in the “in-field” nursery stock treatment for IFA quarantine. However, due to the continued high check mortality in this trial, we propose to duplicate this trial in 2002, to verify results. Prior to the 2002 treatments, we will discuss necessary label changes with the companies involved to determine their current interest in this use pattern for their products.

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- Harlan, D. P., W. A. Banks, H. L. Collins, and C. E. Stringer. 1981. Large area tests of AC217,300 bait for control of imported fire ants in Alabama, Louisiana, and Texas. Southwest. Entomol. 8: 42-45.
- Lofgren, C. S. and D. F. Williams. 1982. Avermectin B_{1a}, a highly potent inhibitor of reproduction by queens of the red imported fire ant. J. Econ. Entomol. 75: 798-803.

Table 1. Bait followed by contact insecticide treatment - Decrease in colony numbers.

Treatment	Mean no. colonies/acre - pretreat	% decrease in no. pretreat colonies at indicated wks. after treatment								
		1*/2**	2*/3**	-4-	-8-	-12-	-17-	-23-	-36-	-44-
Distance + Talstar F*	28.0	100.0a	95.8a	100.0a	100.0a	100.0a	95.2a	100.0a	100.0a	95.2a
Distance + Talstar G**	26.7	95.8a	91.7a	100.0a	100.0a	100.0a	100.0a	100.0a	100.0a	95.2a
Distance + Fipronil G*	28.0	91.1a	100.0a	100.0a	100.0a	96.7a	91.1a	100.0a	96.7a	100.0a
Fipronil Blend**	32.0	90.9a	87.9a	100.0a	100.0a	100.0a	100.0a	97.0a	100.0a	97.0a
Check	38.7	22.1b	17.1b	34.6b	43.3b	51.7b	59.6b	64.2b	70.4b	66.3b

LSD test (P=0.05) means within a column followed by the same letter are not significantly different

Table 2. Bait followed by contact insecticide treatment - Change in population indices.

Treatment	Mean pop. index/acre - pretreat	% change in pretreat population indices at indicated wks. after treatment								
		1*/2**	2*/3**	-4-	-8-	-12-	-17-	-23-	-36-	-44-
Distance + Talstar F*	380.0	-100.0a	-99.4a	-100.0a	-100.0a	-100.0a	-95.5a	-100.0a	-100.0a	-95.5a
Distance + Talstar G**	360.0	-99.4a	-98.4a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a	-95.7a
Distance + Fipronil G*	393.3	-98.4a	-100.0a	-100.0a	-100.0a	-95.2a	-94.1a	-100.0a	-96.4a	-100.0a
Fipronil Blend**	426.7	-92.4a	-92.0a	-100.0a	-100.0a	-100.0a	-100.0a	-96.6a	-100.0a	-97.7a
Check	500.0	4.0b	-6.3b	-9.1b	-29.8b	-42.8b	-52.4b	-59.9b	-65.5b	-59.1b

LSD test (P=0.05) means within a column followed by the same letter are not significantly different

PROJECT NO: A1P04

PROJECT TITLE: Traditional In-field Treatments for Field Grown Nursery Stock: Bait plus
Contact Insecticide, 2002

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon
Wade, Shannon James, Ron Weeks

INTRODUCTION:

Current quarantine treatments for field grown nursery include a bait treatment followed in 3-5 days by a granular chlorpyrifos treatment. This treatment allows for 12 weeks of certification after a 30-day exposure period. With the impending loss of chlorpyrifos, we initiated a trial to replace the chlorpyrifos portion of this treatment regimen with more available chemicals.

MATERIALS AND METHODS:

The bait used was Amdro. The contact insecticides were Talstar 0.2G, Talstar Flowable, and Chipco TopChoice. Rates of application are shown below.

<u>Insecticide</u>	<u>Formulation</u>	<u>Rate of Application</u>
Amdro	bait	1.5 lb/acre
Talstar	0.2G	200 lb/acre (0.4 lb ai/acre)
Talstar	Flowable	40 oz/acre (0.2 lb ai/acre)
fipronil	0.0143G	87 lb/acre (0.0125 lb ai/acre)

The test site was located at the Hattiesburg Municipal Airport in Hattiesburg, MS. As with many airport sites, the upkeep and accessibility is superior, but ant populations tend to be somewhat low. The bait was applied on September 18, 2001 with a shop built applicator mounted on a farm tractor. Air temperature was 85°F and the soil temperature was 83°F. The contact insecticides were applied on September 23-24, 2001 with air temperatures of 79-81°F and soil temperatures of 80°F. Granular material was applied with a Herd™ spreader mounted on a farm tractor. Liquid material was applied with a roller pump boom sprayer equipped with five TKSS tips with provided a 10 ft. swath. The system was operated at 50 psi providing ca. 50 gallons of finished spray per acre. There were three replicates per treatment, and all test plots were 1.0 acre in size. A ¼-acre circular efficacy plot was established in the center of each 1.0 acre test plot. Prior to bait application and at 2, and 4 weeks after final treatment (June 26), IFA populations in each efficacy plot were evaluated using the population index system developed by Harlan et al. (1981), and later revised by Lofgren and Williams (1982). Treatments were evaluated at 4 week intervals thereafter. Using this data, both colony mortality and decrease in pretreatment population indices were calculated. Experimental data were statistically analyzed using analysis

of variance, and treatment means were separated using the LSD test ($P=0.05$) for each posttreatment rating interval.

RESULTS:

Due to adverse weather conditions, the 2 week evaluation was not done. The first evaluation was done at 4 weeks after treatment, at which time all treatments provided 100% control of IFA (Tables 1 and 2). Excellent control continued through the 20 week evaluation. Evaluations will continue until reinfestation is noted.

References Cited:

- Harlan, D. P., W. A. Banks, H. L. Collins, and C. E. Stringer. 1981. Large area tests of AC217,300 bait for control of imported fire ants in Alabama, Louisiana, and Texas. Southwest. Entomol. 8: 42-45.
- Lofgren, C. S. and D. F. Williams. 1982. Avermectin B_{1a}, a highly potent inhibitor of reproduction by queens of the red imported fire ant. J. Econ. Entomol. 75: 798-803.

Table 1. Bait followed by contact insecticide treatment - Decrease in colony numbers.

Treatment	Mean no. colonies/acre - pretreat	% decrease in no. pretreat colonies at indicated wks. after treatment							
		-4-	-9-	-12-	-20-				
Bait + Talstar F	32.0	100.0a	100.0a	100.0a	100.0a				
Bait + Talstar G	33.3	100.0a	100.0a	100.0a	100.0a				
Bait + Fipronil G	29.3	100.0a	100.0a	100.0a	100.0a				
Check	34.7	0.0b	11.8b	7.0b	20.3b				

LSD test (P=0.05) means within a column followed by the same letter are not significantly different

Table 2. Bait followed by contact insecticide treatment - Change in population indices.

Treatment	Mean pop. index/acre - pretreat	% change in pretreat population indices at indicated wks. after treatment							
		-4-	-9-	-12-	-20-				
Bait + Talstar F	422.7	-100.0a	-100.0a	-100.0a	-100.0a				
Bait + Talstar G	418.7	-100.0a	-100.0a	-100.0a	-100.0a				
Bait + Fipronil G	460.0	-100.0a	-100.0a	-100.0a	-100.0a				
Check	437.3	56.2b	-1.7b	-1.6b	-44.8b				

LSD test (P=0.05) means within a column followed by the same letter are not significantly different

PROJECT NO: GPPS01-04

PROJECT TITLE: Evaluation of Talstar™ For Control of Imported Fire Ants in Turf Grass, 2001

TYPE REPORT: Final

LEADERS/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade and Chris Doxey

INTRODUCTION:

Talstar (bifenthrin), in both granular and liquid formulations, have been evaluated by this laboratory for control of IFA in grass sod. Results with the granular formulation has been somewhat variable (FA01G063, FA01G066, FA01G028), but excellent control was obtained in several trials with the flowable formulation (FA01G065, FA01G066, GPPS00-06).

MATERIALS AND METHODS:

The test site was located at the Slidell Municipal airport in Slidell, LA. Test plots were one acre in size, with a ¼ acre efficacy subplot located in the center of the test plots. Liquid bifenthrin were applied with a roller pump boom sprayer equipped with five TKSS tips with provided a 10 ft. swath. The system was operated at 50 psi providing 38 gallons of finished spray per acre. Rates of application included a 0.1 lb ai/acre dual application (0.1 lb ai/acre applied twice, one week apart), and single applications of 0.2 and 0.4 lb ai/acre. The first of the dual applications was made on May 17, 2001. The second dual application and the single applications were made on May 21 and 22, 2001. Prior to treatment and at 1, 2, and 4 weeks after treatment evaluations of IFA populations were made in each ¼ acre efficacy subplot using the procedures described by Lofgren and Williams (1982) and Collins and Callcott (1995). Evaluations were made at 4 week intervals thereafter. Differences in treatment means were separated by a LSD test (P=0.05).

RESULTS:

In the 6-8 weeks prior to our treatment and continuing into the first evaluation, the treatment site had not received much rainfall (if any). At one week after treatment, there were decreases in colony numbers and population indices, however, the differences were not significantly different than the untreated check (Table 1 and 2).

Due to the rains of Tropical Storm Allison June 6-11, 2001, we were not able to perform a 2 or 3 week evaluation. During this time the treatment site received more than 18 inches of rainfall.

At 4 weeks after treatment, all treatments provided >90% control, with the split application and the high single application providing the best numerical control. By 8 weeks, the split application and the high single rate still provided the best numerical control with 97-100%

control of IFA in grass sod. High control mortality was noted at 8 weeks. This is not unusual for south Mississippi during late July and August, which are usually very hot and dry. Prior to the 13 week evaluation, the site had +15.0 inches of rainfall, mostly during the 2 weeks prior to the evaluation. At that time, all treatments were significantly better than the check. However, only the 0.4 lb/acre rate provided 100% control. The two lower rates provided similar numerical control at ca. 90%. At 17 weeks, some reinfestation was noted on the two lower rates of application, while the 0.4 lb ai/acre rate maintained 100% control. At 20 weeks, reinfestation of the two low rates continued and evaluations of these plots was discontinued. The 0.4 lb ai/acre rate continued to provide good control through 33 weeks, with some decrease in efficacy at 41 weeks.

Recommendation:

Current Talstar labelled rates for treatment of IFA on sod farms is 0.2 lb ai/acre. Unfortunately, this rate of application does not provide consistent rates of mortality or effective residual activity adequate for regulatory/quarantine use. The 0.4 lb ai/acre rate is much more consistent, faster acting and provides an effective residual activity. In discussions with the company, they are reluctant to increase the label rate of application, but are interesting in developing an acceptable quarantine treatment for IFA on grass sod. In the past, this laboratory did one trial in which Talstar liquid was applied at a rate of 0.2 lb ai/acre per application, with two applications one week apart. In that trial (FA01G066), 100% control was achieved by 12 weeks after treatment, and maintained through 28 weeks (trial terminated due to harvest). Therefore, in 2002, we propose to initiate trials at two to three locations in the southern Mississippi area, that re-evaluate the efficacy of the multiple treatments of Talstar liquid applied at 0.2 lb ai/acre per application, with two applications being made approximately one week apart.

REFERENCES CITED

- Collins, H. L. and A.-M. A. Callcott. 1995. Effectiveness of spot insecticide treatments for red imported fire ant (Hymenoptera: Formicidae) control. J. Entomol. Sci. 30: 489-496.
- Lofgren, C. S. and D. F. Williams. 1982. Avermectin B_{1a}, a highly potent inhibitor of reproduction by queens of the red imported fire ant. Jour. Econ. Entomol. 75: 798-803.

Table 1. Efficacy of Talstar grass sod treatments - Decrease in colony numbers.

Treatment: lb ai/ acre	Mean no. col./acre – pretreat	% decrease in no. pretreat colonies at indicated wks. after treatment								
		-1-	-4-	-8-	-13-	-17-	-20-	-24*-	-33-	-41-
Talstar F: 0.1+0.1	54.7	53.0a	94.3a	100.0a	90.0a	84.3a	64.3ab	--	--	--
Talstar F: 0.2	57.3	36.4a	90.7a	87.2a	90.3a	86.1a	47.3ab	--	--	--
Talstar F: 0.4	57.3	59.7a	97.2a	97.2a	100.0a	100.0a	97.2a	92.8a	95.0a	95.0a
Check	34.7	33.3a	45.0b	65.0b	52.5b	41.7b	12.5b	8.3b	53.3b	26.7b

LSD test (P=0.05); means within a column followed by the same letter are not significantly different

* t-test (P=0.05) from this point on

Table 2. Efficacy of Talstar grass sod treatments - Change in population indices.

Treatment: lb ai/ acre	Mean pop. index/ acre - pretreat	% decrease in pretreat population indices at indicated wks. after treatment								
		-1-	-4-	-8-	-13-	-17-	-21-	-24*-	-33-	-41-
Talstar F: 0.1+0.1	960.0	-62.8a	-99.2a	-100.0a	-91.4a	-86.4a	-71.4ab	--	--	--
Talstar F: 0.2	946.7	-43.5a	-99.5a	-90.3a	-91.6a	-86.1a	-64.2ab	--	--	--
Talstar F: 0.4	966.7	-63.6a	-96.2a	-97.3a	-100.0a	-100.0a	-97.6a	-95.0a	-95.0a	-87.7a
Check	593.3	-43.1a	-52.3b	-67.9b	-58.8b	-44.2b	-27.6b	-20.9b	-61.1b	-35.9b

LSD test (P=0.05); means within a column followed by the same letter are not significantly different

* t-test (P=0.05) from this point on

PROJECT NO: GPPS02-05

PROJECT TITLE: New Treatments for Control of Imported Fire Ants on Grass Sod, 2002

TYPE REPORT: Final

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade

INTRODUCTION:

Currently, there is only one quarantine treatment for control of IFA on commercial grass sod and only one label that can be used. Dursban 50W is applied at 8 lb ai/acre, and after a 48 hour exposure, there is a 6 week certification period. Aventis Crop Science (Bayer) has obtained a label for fipronil and that label does include a grass sod treatment. However, APHIS approval for this product is not expected before spring 2002. Regardless of this, we need more options for the sod growers shipping outside the quarantined area. Talstar (bifenthrin), in both granular and liquid formulations, have been evaluated by this laboratory for control of IFA in grass sod. Results with the granular formulation has been somewhat variable, but excellent control was obtained in several trials with the flowable formulation.

MATERIALS AND METHODS:

Two sites in Forrest Co., Mississippi were selected; one at the Hattiesburg Municipal Airport and the other at the Camp Shelby National Guard Base. The airport site is very similar to a grass sod in height of grass, moving frequency, although grass type is mixed and abundance is less than a sod farm. The Camp Shelby site is more of a natural pasture site, with less frequent moving, taller grass and mixed grasses. Neither site has supplemental irrigation (not all sod farms have supplemental irrigation). Test plots were one acre in size with a ¼ acre efficacy subplot located in the center of the test plots. Liquid bifenthrin was applied with a roller pump boom sprayer equipped with five flood jet tips (1/8 KSS 7.5) and provided a 10 ft. swath with approximately 55 gallons of finished liquid being applied per acre. Rates of application were 0.2 lb ai/acre dual application (0.2 lb ai/acre applied twice, one week apart). First treatments were applied on April 17 and 18, 2002 with the second applications made on April 23 and 24, 2002. Air temperatures range from 78 to 83°F on treatment days and soil temperatures ranged from 68 to 72°F.

Prior to treatment and at 2 and 4 weeks after treatment evaluations of IFA populations were made in each ¼ acre efficacy subplot using the procedures described by Lofgren and Williams (1982) and Collins and Callcott (1995). Evaluations were made at monthly intervals thereafter. Differences in treatment means were separated by a t-test.

RESULTS:

There was no rainfall 1 week prior to treatment, no rainfall between treatments, and no rainfall prior to the first evaluation at 2 weeks after the last treatment. It was also unseasonably warm during this period (ca. 90°F every day).

Results are shown by site and combined (Tables 1-6). Treatments provided greater than 90% control of IFA at both sites by 4 weeks after treatment. The airport site achieved 100% mortality at 8 weeks while the Camp Shelby site achieved 100% mortality at 12 weeks after treatment. Both maintained excellent control through 20 weeks, although the Camp Shelby site did have a slight decrease from 100% control. Detectable presence of IFA on control sites was greatly reduced during the early counts (particularly 4-12 week counts) due to early extreme heat and drought. These conditions are normally not seen until late July and August. Tall grass on the Camp Shelby site also impeded visual evaluations.

Excellent control was obtained at both sites. The Camp Shelby site was not an ideal test site due to the un-managed nature of the site. Grass was much longer at application and was not mowed until very late in the evaluation period. The airport site was regularly mowed and the grass was much shorter at application. The differences in these sites indicate better control at the site with shorter grass. The airport site is much more indicative of a commercial sod farm. In a previous trial (FA01G066 – 1996), the dual application of bifenthrin flowable on grass sod provided 100% control 12 weeks after application, and maintained that level of control through 28 weeks (16 weeks after reaching 100%). In another trial (GPPS01-04), a single 0.4 lb ai/acre rate achieved 100% control at 13 weeks, and sustained that control through 17 weeks (97% control at 20 weeks). In the current trial, 100% control was achieved at the airport site at 8 weeks, and maintained through 20 weeks (12 weeks after reaching 100%). Additional trials will be necessary to firmly establish time to achieve 100% control and length of control.

Table 1. Hattiesburg: % colony kill

Treatment	Mean no. mounds/acre – pretreat	Percent decrease in no. pretreat colonies at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	34.7	83.3a	91.7a	100.0a	100.0a	100.0a	100.0a	73.8a
Check	38.7	42.8a	60.2a	54.6a	28.3b	22.2b	12.5b	4.2b
P value		0.145	0.162	0.113	0.012	0.034	0.020	0.033

Table 2. Camp Shelby: % colony kill

Treatment	Mean no. mounds/acre – pretreat	Percent decrease in no. pretreat colonies at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	69.3	87.0a	92.1a	90.7a	100.0a	100.0a	98.6a	93.0a
Check	61.3	57.8a	51.3b	39.0b	64.5b	62.8b	56.8b	25.4b
P value		0.088	0.030	0.006	0.046	0.019	0.010	<0.001

Table 3. Hattiesburg: % change in population index

Treatment	Mean pop. index/acre – pretreat	Percent change in pretreat population indices at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	560.0	-89.6a	-95.1a	-100.0a	-100.0a	-100.0a	-100.0a	-84.6a
Check	600.0	-53.0a	-67.3a	-66.0a	-33.5a	-21.5b	-17.9b	24.4b
P value		0.077	0.122	0.112	0.087	0.047	0.038	0.005

Table 4. Camp Shelby: % change in population index

Treatment	Mean pop. index/acre – pretreat	Percent decrease in pretreat population indices at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	1266.7	-91.1a	-97.2a	-99.0a	-100.0a	-100.0a	-99.9a	-95.2a
Check	1186.7	-66.6a	-63.7b	-50.8b	-71.5a	-69.5b	-65.8b	-35.8b
P value		0.066	0.046	0.047	0.056	0.016	0.012	0.011

Table 5. Combined data: % colony kill.

Treatment	Mean no. mounds/acre – pretreat	Percent decrease in no. pretreat colonies at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	52.0	85.1a	91.9a	95.4a	100.0a	100.0a	99.0a	83.4a
Check	50.0	50.3b	55.8b	46.8b	46.4b	42.5a	34.7b	14.8b
P value		0.016	0.006	<0.001	0.005	0.004	0.003	<0.001

Table 6. Combined data: % change in population index

Treatment	Mean pop. index/acre – pretreat	Percent decrease in pretreat population indices at indicated wks posttreat						
		-2-	-4-	-8-	-12-	-16-	-20-	-26-
Talstar F	913.3	-90.3a	-96.1a	-99.5a	-100.0a	-100.0a	-99.9a	-89.9a
Check	893.3	-59.8b	-65.5b	-58.4b	-52.5b	-45.5b	-41.8b	-5.7b
P value		0.006	0.006	0.004	0.015	0.010	0.007	0.003

PROJECT NO: A1P03

PROJECT TITLE: Evaluation of Various Chemical and Physical Barriers to Preclude Invasion of Rolled Hay Bales Stored in the Field, 2002

TYPE REPORT: Final

LEADER/PARTICIPANTS: Timothy C. Lockley

INTRODUCTION:

One of the products currently listed under federal quarantine as a possible vector for shipment of imported fire ants is baled hay and straw. No economically viable methods exist to treat bales exposed to fire ant invasion and currently only baled hay and straw not stored in direct contact with the ground may be moved.

MATERIALS AND METHODS:

Evaluations of potential barriers to fire ant movement into rolled hay bales were made at the White Sands farm of the Mississippi Agriculture and Forestry Experiment Station in Pearl River County. In all trials, hay was baled in one field and removed within 24 hours to another field in which the trials were to be conducted; however bale placement on actual treatment site occurred within 24-72 hours of baling. For example, in the first trial, bales were left in the “baling” field for 24 hours and then moved directly to the test field and placed on actual treatment sites. In the other trials, bales were removed from the baling field immediately after baling and “stored” in the test field until actual treatment sites were ready (baling late Friday afternoon resulted in bales not being placed on treated areas until Monday morning).

Experiment 1 began on 24 May, 2002 and continued through August of that year. Rolled bales of hay were placed ca. 7 m apart on a grid pattern in a 2.25 ha field actively infested with monogyne colonies of the red imported fire ant, *Solenopsis invicta* Buren. Materials tested consisted of:

- Check
- Diazinon 25 %
- Malathion 50%
- Large Truck Tires

Three replicates of each candidate plus a check were set up in a randomized block. Chemical treatments were broadcast over a 5 x 5 m area. Tires were placed in the middle of a 5 x 5 m square. Bales were set in the middle of each treatment. Bales were set upon the tires in such a manner as to preclude their touching the ground. Examinations of each bale for the presence of fire ant colonies were made each month.

A second experiment was begun on 9 August, 2002. Chemical treatments to the ground followed the protocol described in Experiment 1. Prefabricated wooden pallets were treated 24 hrs prior to placement in the field. Treatments consisted of:

- Check
- Dylox 6.2G ground treatment
- Lannate 90WP ground treatment
- Malathion 50% pallet treatment
- Diazinon 25% pallet treatment

A third experiment was initiated on 5 November, 2002. Treatments consisted of elevating bales on untreated tires and pallets.

RESULTS:

The outcomes of each experiment are shown in the tables below. All treatments, except the Dylox soil treatment, appeared to preclude IFA colonies from infesting bales for a minimum of 2 months after placement. Additional trials will be conducted in 2003 to replicate the promising treatment options and continue to explore better methods of excluding IFA from baled hay.

Table 1. Efficacy of various physical and chemical barriers to exclude imported fire ant colonies from rolled hay bales.

Candidate	No. infested bales/ reps at indicated mths PT		
	(1)	(2)	(3)
Tire	0	0	0
Diazinon	0	0	1
Malathion	0	0	1
Check	0	2	2

Table 2: Efficacy of various chemical and chemically treated barriers to exclude imported fire ant colonies from rolled hay bales.

Candidate	No. infested bales/rep at indicated mths PT			
	(1)	(2)	(3)	(4)
Pallet (Diazinon)	0	0	1	2
Pallet (Malathion)	0	0	0	1
Dylox	1	1	2	3
Lannate	0	0	2	1
Check	0	1	1	2

Table 3. Efficacy of two untreated mechanical barriers to exclude imported fire ant colonies from rolled hay bales.

Candidate	No. infested bales/rep at indicated mths PT		
	(1)	(2)	(3)
Pallet	0	0	0
Tire	0	0	0
Check	2	3	3

PROJECT NO: A1P03

PROJECT TITLE: Observations of Red Imported Fire Ants In and Around Freshly Baled Hay, 2002

REPORT TYPE: Final

PROJECT LEADER/PARTICIPANTS: Shannon Wade

INTRODUCTION:

One of the products currently listed under the federal quarantine as a possible vector for shipment of imported fire ants is baled hay and straw. No economically viable methods exist to treat bales exposed to fire ant invasion. Currently, only baled hay and straw not stored in direct contact with the ground may be moved.

MATERIALS AND METHODS:

Observations of fire ants in and around freshly baled hay were made at the Upton David farm in Jackson County, Mississippi.

On September 6, 2002 hay was square baled around 2:30pm without rain. Most of the square bales had workers, but no brood, in them immediately after being baled. Fifteen bales were left in the field for further observations while the rest were moved into a hay loft. Three of the bales left in the field did not have ants in them immediately after baling.

OBSERVATIONS:

Day 1-The bales of hay were not examined the day after being baled because of 1 inch of rain.

Day 2-The bales were examined around 7:30pm. The bales were very wet on the inside, but outside had dried out. The ground was dry around the bales but very wet underneath. The three bales that started out with no ants in them still had no ants in them. The other 12 bales that originally had ants in them now had colonies in them. Most of the colonies were underneath the bales with lots of workers and brood. Three of the bales had two separate colonies under the bales and built up at both ends. The rest of the infested bales just had one colony of ants, either under the bale or built up on the sides or end.

Day 5- Bales of hay were again examined around 7:30pm. Of the three bales that previously were not infested, one had a small colony move in under the bale. The other two had about 100 workers under and in the bale, possibly moving in or foraging. Seven of the bales were still the same as they were on Day 2. One bale had a mound that increased in size. The other four infested bales had colonies that appeared to be decreasing in size.

Day 14- Again bales were not examined because of 2 ½ inches of rain.

Day 15- Bales of hay were very wet, along with the ground underneath. Seven of the previously infested bales were still the same. Only one bale, of the three bales that started off with no ants, and then had a few by Day 5, still had one colony under. Three bales that were originally infested no longer had any ants present. And, the last two only had a small amount of workers underneath.

CONCLUSION:

After a couple of months of storage, bales that were moved to a hay loft immediately after baling, posed no problems to persons handling the bales or to animals consuming them. This indicates that any workers that were baled up with the hay and moved into the loft had left the bales.

At least for a short period of time, leaving the bales in the immediate area in which the IFA workers were baled into the hay bales seemed to encourage the workers to relocate the rest of the colony into the bales. Also, the rain appears to have created a nice moist environment, enticing the ants to stay or move in.

While 80% of the bales had colonies present 2 days after baling, only 53% of the bales had colonies in or under them at the end of the two week period. And, 13% of the bales had workers only.

Further observations are still necessary in this area.

PROJECT NO: A1P02

PROJECT TITLE: Evaluation of New Imported Fire Ant Bait Formulations, 2002

TYPE REPORT: Final

LEADER/PARTICIPANTS: Anne-Marie Callcott, Lee McAnally, Tim Lockley, Shannon Wade

INTRODUCTION:

Imported fire ant baits are an important tool for the homeowner battle against IFA, as well as being used in the Federal IFA Quarantine (7CFR 301.81). While baits generally do not provide 100% control when used alone against IFA, nor do they provide residual activity due to the inherent nature of baits, they are becoming more important as an environmentally friendly tool in an integrated management approach to controlling IFA. BASF Corp. (RTP, NC) and Dupont Co. (Newark, DE) each provided us with one active ingredient formulated on a bait carrier at various dose rates. Both products were tested in the laboratory against either field collected or lab-reared IFA colonies. Promising results indicated that field testing of both baits was warranted.

MATERIALS AND METHODS:

The test site was located on open pastureland off Landon Rd, in Gulfport, Mississippi (Fore property). Baits were applied on June 11, 2002 with air temperature at 84-88°F and soil temperature at 75-79°F. All bait formulations were applied with a shop-built granular applicator mounted on a farm tractor. The equipment provided a 21' swath and was operated at 4 mph. Each time a different formulation was applied the equipment was re-calibrated to deliver ca. 1.5 lbs of bait per acre. Actual application was closer to 1.0 lb/acre. There were three replicates per treatment, and all test plots were 1.0 acre in size. A ¼-acre circular efficacy plot was established in the center of each 1.0 acre test plot. Prior to bait application and at 4 week intervals, IFA populations in each efficacy plot were evaluated using the population index system developed by Harlan et al. (1981), and later revised by Lofgren and Williams (1982). Using this data, both colony mortality and decrease in pretreatment population indices were calculated. Experimental data were statistically analyzed using analysis of variance, and treatment means were separated using the LSD test ($P=0.05$) for each posttreatment rating interval.

RESULTS:

Little rainfall occurred between treatment and the 4 week evaluation, while significant rainfall occurred between the 4 and 8 week evaluation period (+12 inches during 2 main rain events – tropical storm/hurricane), and rain regularly fell through the 16 week evaluation.

At 4 weeks after treatment, both baits showed dose dependent activity, with the highest amount of active ingredient providing the greatest control (Tables 1-6). Efficacy remained generally dose dependent through out the trial; the higher dose rates being more effective. The BASF bait did reduce population indices significantly compared to the untreated checks at weeks 8 and 12

(Table 4), but decrease in colony numbers (Table 3) was numerically low (19-74% reduction in colony numbers). The Dupont bait was numerically superior to the BASF bait in reducing colony numbers, but never achieved better than 91% colony reduction and generally was in the 70% reduction range (Table 5). This bait did provide >87% decrease in population indices at weeks 4-12 (Table 6). Both baits at all rates were showing significant reductions in control by 16 weeks, which is fairly typical of baits that have no residual activity, and thus the trial terminated.

Table 1. Decrease in colony numbers – all baits.

Treatment	Mean no. colonies/acre – pretreat	Percent decrease in pretreat colony numbers at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
BASF 0.063%	36.0	37.8d	27.3bc	59.4bc	44.4ab	-
BASF 0.125%	40.0	46.8cd	19.0c	41.0cd	33.3ab	-
BASF 0.25%	36.0	65.3bc	47.1abc	73.9ab	56.2a	-
Dupont 0.025%	40.0	74.4ab	60.0ab	75.6ab	41.1ab	-
Dupont 0.05%	33.2	83.0ab	64.8ab	87.4a	49.2ab	-
Dupont 0.1%	36.0	91.4a	73.6a	79.5ab	57.9a	-
Check	34.8	34.7d	19.9c	19.4d	11.1b	-

Means within a column followed by the same letter are not significantly different (LSD test, $P<0.05$)

Table 2. Change in population indices – all baits.

Treatment	Mean pop. index/acre – pretreat	Percent change in pretreat population indices at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
BASF 0.063%	626.8	-73.2bcd	-75.6bc	-76.9a	-46.9ab	-
BASF 0.125%	706.8	-72.3cd	-69.4c	-77.6a	-45.1ab	-
BASF 0.25%	640.0	-94.6ab	-85.3ab	-85.3a	-70.2a	-
Dupont 0.025%	766.8	-92.0abc	-92.3a	-87.4a	-44.1ab	-
Dupont 0.05%	620.0	-97.3a	-87.0ab	-92.4a	-54.3ab	-
Dupont 0.1%	640.0	-96.8a	-95.7a	-96.9a	-70.5a	-
Check	660.0	-51.7d	-38.2d	-18.9b	-11.6b	-

Means within a column followed by the same letter are not significantly different (LSD test, $P<0.05$)

Table 3. Decrease in colony numbers – BASF.

Treatment	Mean no. colonies/acre – pretreat	Percent decrease in pretreat colony numbers at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
BASF 0.063%	36.0	37.8ab	27.3a	59.4ab	44.4a	-
BASF 0.125%	40.0	46.8ab	19.0a	41.0bc	33.3a	-
BASF 0.25%	36.0	65.3a	47.1a	73.9a	56.2a	-
Check	34.8	34.7b	19.9a	19.4c	11.1a	-

Means within a column followed by the same letter are not significantly different (LSD test, $P < 0.05$)

Table 4. Change in population indices – BASF.

Treatment	Mean pop. index/acre – pretreat	Percent change in pretreat population indices at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
BASF 0.063%	626.8	-73.2ab	-75.6a	-76.9a	-46.9ab	-
BASF 0.125%	706.8	-72.3ab	-69.4a	-77.6a	-45.1ab	-
BASF 0.25%	640.0	-94.6a	-85.3a	-85.3a	-70.2a	-
Check	660.0	-51.7b	-38.2b	-18.9b	-11.6b	-

Means within a column followed by the same letter are not significantly different (LSD test, $P < 0.05$)

Table 5. Decrease in colony numbers – Dupont.

Treatment	Mean no. colonies/acre – pretreat	Percent decrease in pretreat colony numbers at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
Dupont 0.025%	40.0	74.4a	60.0a	75.6a	41.1ab	-
Dupont 0.05%	33.2	83.0a	67.8a	79.5a	49.2ab	-
Dupont 0.1%	36.0	91.4a	73.6a	87.4a	57.9a	-
Check	34.8	34.7b	19.9b	19.4b	11.1b	-

Means within a column followed by the same letter are not significantly different (LSD test, $P < 0.05$)

Table 6. Change in population indices – Dupont.

Treatment	Mean pop. index/acre – pretreat	Percent change in pretreat population indices at indicated weeks after treatment				
		-4-	-8-	-12-	-16-	-20-
Dupont 0.025%	766.8	-92.0a	-92.3a	-87.4a	-44.1a	-
Dupont 0.05%	620.0	-97.3a	-87.0a	-92.4a	-54.3a	-
Dupont 0.1%	640.0	-96.8a	-95.7a	-96.9a	-70.5a	-
Check	660.0	-51.7b	-38.2b	-18.9b	-11.6a	-

Means within a column followed by the same letter are not significantly different (LSD test, $P < 0.05$)

PROJECT NO: A9P03

PROJECT TITLE: Mississippi Phorid Fly Release Project

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Timothy Lockley; Sanford Porter (USDA, ARS), Shannon James

INTRODUCTION:

Pseudacteon species are endoparasites of *Solenopsis* species and are widely distributed throughout the fire ant range in their natural habitats. These phorid flies have a potential to suppress fire ant populations if they can become established in North America. To determine their ability to acclimatize, these phorids were released in the spring of 2000 in Harrison County, MS.

MATERIALS AND METHODS:

Release I: A release site near Saucier and a paired control site at the Harrison County Work Farm were selected for the study. The sites were ca. 20 km apart. Each site had similar habitats at the time of release; consisting of grassland with deciduous woods and large ponds adjacent. Both the release site and the control site had ca. 100 active monogyne colonies of IFA per ha. Emerged adult flies of *Pseudacteon tricuspsis*, supplied by S. Porterm were released daily, per the protocol provided by S. Porter, at the Saucier site beginning on 11 April, 2000 with the final release occurring on 20 April. A total of 2612 phorids were released on 45 separate imported fire ant colonies.

Release II: In August 2002, over 2000 phorids were released at the Hattiesburg Airport (Bobby Chain) as a part of the APHIS phorid fly rearing and release project. Evaluations on phorid establishment will be made beginning in the spring of 2003.

RESULTS:

Release I: The first survey was completed in July 2000, 3 months after release. Phorids were observed at each of five excavated mounds within the immediate confines of the release site. No flies were observed outside of the field. Another survey accomplished in October 2000 again revealed the presence of phorids within the release field but none were found outside of the site.

In April 2001, one year after release, flies were seen ca. 200 m from the initial point of release. The release site was converted from cattle-grazed pastureland to a pine tree farm in 2001 and no successful surveys could be conducted at the survey site in the fall of 2001. A search of the roadside area adjacent to the release site, however, showed phorids. In October 2001, phorids were observed ca. 3.5 km south of the original release point.

An attempt to evaluate survival of phorids and their range expansion in May 2002 failed to show any activity even at the original release site. A 5 week period of unseasonably hot weather with no rainfall may have been the cause of the failure to detect flies. In October 2002, a survey along the cardinal points showed a marked movement by the flies. To the south, movement of ca. 8 km was noted. To the west, phorids were observed at ca. 10 km distant from the original release point. To the north and east, flies were detected at ca. 4 km.

Release II: A preliminary evaluation in November 2002 showed the presence of flies at the release site indicating establishment successive generations of the original flies. Overwinter evaluations have not been made to date.

PROJECT NO: A9P03 - FA02G049

PROJECT TITLE: Evaluation of Field Releases of *Thelohanian solenopsae*, 1999-2002

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Anne-Marie Callcott, Shannon James, Shannon Wade, Lee McAnally, Tim Lockley, Homer Collins, and Avel Ladner

COOPERATORS: Drs. David Williams and David Oi, USDA, ARS, CMAVE, Gainesville, FL

INTRODUCTION:

The microsporidium *Thelohanian solenopsae* (Microsporidia: Thelohaniidae) was discovered in Brazil in the red imported fire ant (Knell et al. 1977). Since that time, USDA, ARS, CMAVE personnel in Argentina have also discovered the pathogen in the black imported fire ant in that country and have determined that the pathogen does decrease colonies and colony vigor and therefore may be a good candidate for use as a biological control agent in the United States (Briano et al. 1995a, 1995b, 1996). In 1998, we initiated a trial releasing the microsporidium in Harrison and Hancock counties, MS (FA02G048). These initial inoculation sites were lost or had poor results. Therefore, we repeated the trial in the fall of 1999.

MATERIALS AND METHODS:

In October, 1999 we assisted ARS with the initiation of a trial to evaluate field releases of the pathogen *Thelohanian solenopsae*. Two sites, one polygyne in Hancock Co. and one monogyne in Harrison Co., were selected for the inoculation and four plots set up at each site. At the polygyne site, circular test plot evaluation areas were $\frac{1}{16}$ acre in size due to the large number of mounds in the area. Two plots were used as inoculation plots and two were maintained as non-inoculated control plots. On October 19, 1999 nine mounds in each of the inoculation plots were inoculated with 3.5g of brood infected with *T. solenopsae* (field collected by ARS prior to study). At the monogyne site, circular test plot evaluation areas were the standard $\frac{1}{4}$ acre size. Inoculations were also made on October 19 to nine mounds in each of two test plots. Every two months we monitor the inoculated plots and corresponding non-inoculated control plots by evaluating mounds with the mound index system, geo-referencing each mound within the plots, and collecting worker samples from each mound within the plots. We also assist by microscopically examining collected workers for pathogen spores.

RESULTS:

Colony mortality

Small decreases in number of colonies present in both the monogyne and polygyne sites were seen with the first post-inoculation evaluation (Table 1). Since decreases also occurred in the control plots, this probably cannot, at this time, be attributed to the pathogen. Decreases in population indices were more significant, particularly in the monogyne site. However, these

decreases were mainly due to many colonies not having worker pupae present. This may be a seasonal trend (brood production slows down in the winter months). In the monogyne site, the control plots on average maintained colony numbers and populations through 49 weeks (11 months), while the inoculated plots showed decreases in both (Tables 1 & 2). The monogyne plots were lost after the 49 week evaluation due to pasture improvements.

Through the 155 weeks (3 years) of the trial thus far, the polygyne site has shown fluctuation in both colony numbers and population indices. This fluctuation, however, appears to be seasonally cyclical. Significant decreases in colony numbers and population indices occurred in the control plots and at weeks 49, 92, and 141 during the dry heat of summer, when ants would be deeper in the mounds to conserve moisture. Likewise, populations appeared to peak annually in spring samples at 28, 76, and 127 weeks. Through week 76 the inoculated plots maintained similar colony number and population index readings as the controls echoing their peaks and drops. However, at week 84 and thereafter the population index for inoculated colonies remained at less than half the pretreatment reading while the control displayed population readings from almost half the initial reading in the summer heat of week 92 to a 9 percent increase over initial readings in the spring sample of week 127. Growth of colony number displayed a similar difference between the control and inoculated plots during this span, indicating some long-term effects of the pathogen. While differences in colony number and population index are evident numerically from week 84 through 127, with only two replicates, statistical analysis is suspect, and therefore was not done.

Between our readings at weeks 141 and 155, rains and high waters from tropical storm Isadore and hurricane Lili flooded our test site. Very few colonies were located in the area after this flooding. As of week 161 the number of control plot colonies had not returned to the level expected for that time of year. We will continue monitoring at least through 2003, but it is possible that the floods during fall of 2002 have disrupted this site beyond further use.

Presence of pathogen

Pretreatment samples were examined and no spores were detected at either site. At 12 weeks after inoculation (January 2000), 2 mounds in one of the inoculated polygyne plots were positive for spores. By week 20, both of the inoculated polygyne plots had three positive colonies, and a few spores were detected from 2 polygyne control plot mounds. Since that sample, no other spore positive results have been found for the control plots. No spores were detected in the monogyne site through 20 weeks, and no other monogyne samples were evaluated thereafter. The percent of infected colonies in both inoculated polygyne plots increased as the number of colonies decreased (Table 3). While one of these inoculated plots only had one or two positive colonies per sample date, after the three found at week 20, the other plot has shown progression with a maximum of four infected colonies in a sample date by year two and eight by year three. There does seem to be some fluctuation with observation of infection as colony samples will test positive on one date and not the next and then positive again later on. This may be due to colony movement. The period of highest incident of colony infection falls within the sample dates when the inoculated plots consistently had far lower colony numbers and population indices than the control plots, indicating that infection was probably responsible. The polygyne site will continue to be monitored for *Thelohania solenopsae* infection for at least the next year.

All data generated by this trial will be compiled and reported on in full by the USDA, ARS, CMAVE cooperators in Gainesville, FL. They have duplicated this trial in several different geographical locations and will be responsible for data summary and publication.

References Cited:

- Briano, J., R. Patterson and H. Cordo. 1995a. Relationship between colony size of *Solenopsis richteri* (Hymenoptera: Formicidae) and infection with *Thelohania solenopsae* (Microsporidae: Thelohaniidae) in Argentina. J. Econ. Entomol. 88: 1233-1237.
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Table 1. Mean percent change in colony number in *Thelohania solenopsae* inoculated sites – inoculated October, 1999.

Weeks post inoculation	Polygyne		Monogyne	
	Inoculated	Control	Inoculated	Control
12	-12.8	-17.9	-5.5	40.0
20	-2.5	-8.0	-2.8	42.9
28	8.1	-12.9	-2.6	57.2
36	-12.5	-40.6	-17.7	5.0
49	-66.1	-43.4	-27.4	55.0
76	10.5	5.7	-	-
84	-42.5	-21.7	-	-
92	-55.3	-38.1	-	-
100	-42.1	-23.8	-	-
109	-47.4	-9.5	-	-
118	-50.3	-25.8	-	-
127	-52.8	-3.2	-	-
141	-60.3	-54.4	-	-
155	-86.7	-63.7	-	-
161	-63.1	-61.7	-	-
Mean no. colonies/acre pretreat	304	336	66	34

Table 2. Mean percent change in population index in *Thelohania solenopsae* inoculated sites – inoculated October, 1999.

Weeks post inoculation	Polygyne		Monogyne	
	Inoculated	Control	Inoculated	Control
12	-28.9	-17.3	-53.7	-13.5
20	-5.9	-10.4	-7.6	50.0
28	2.3	-0.7	0.4	65.8
36	-32.9	-50.7	-36.5	-0.1
49	-76.1	-54.9	-32.5	30.9
76	-5.7	4.2	-	-
84	-69.4	-30.6	-	-
92	-62.2	-47.5	-	-
100	-56.8	-30.8	-	-
109	-52.3	-10.0	-	-
118	-58.9	-27.9	-	-
127	-55.1	9.1	-	-
141	-66.7	-59.8	-	-
155	-83.6	-61.8	-	-
Mean pop. index/acre pretreat	4,440	4,800	1,160	590

Table 3. Mean percent of infected colonies in the polygyne site inoculated with *Thelohania solenopsae* October, 1999.

Weeks post inoculation	Inoculated*	Control*
12	6.1	0
20	16.3	5.9
28	2.5	0
36	3.4	0
49	12.5	0
76	9.6	0
84	-	-
92	12.5	0
100	19.7	0
109	22.6	0
118	48.9	0
127	52.5	0
141	7.1	0
155	33.4	0
161	35.7	0

* Information for samples collected for week 84 is unavailable.

PROJECT NO: A2P01

PROJECT TITLE: Test and Develop Universally Acceptable Fire Ant Bait for Survey Traps –
Part I: Tests of Commercially Available Products for Use as IFA Survey Baits.

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Robert G. Jones, Ph.D, Timothy Lockley

INTRODUCTION:

Numerous baits are used to attract the imported fire ant (IFA) for both survey and research studies. There is no consistency in what is being used among the states. The use of many are laborious, messy or both. While attractants have been evaluated over the years, most researchers have not determined that one bait attractant is superior over others for use in survey traps (Lofgren et al. 1961, Brinkman et al. 2001). There are some bait attractants currently in use that have not been compared for effectiveness. None have been tested for attractiveness to all three types of imported fire ants; the red, black and hybrid.

Laboratory testing was done to screen out the most promising products from the less promising. The more promising “off the shelf” products were then tested in the field.

METHODS AND MATERIALS:

Laboratory tests, conducted March-June 2002, were set up comparing numerous commercial products (Table 1) versus a standard bait of peanut cooking oil (30%) on pregelled corn carrier (70%). The test method was a standardized Gulfport IFA Laboratory protocol based on Lofgren et al. (1961). Field collected worker ants (adult and immature) with mound soil were collected with a small bladed shovel and placed in a plastic 11.4 quart dishpan or 12 qt. sweater box. Each treatment was composed of 5 replications of collected ants in plastic boxes. The insides of the plastic boxes were dusted with talcum powder to prevent escape. The ants were held for 3 to 5 days before testing. On the day of the test the soil was watered and a board (1’x2’x12”) was placed in the box on the soil. At each end of the board a petri dish (100mm x 15mm square or round) was placed with one dish bottom containing 4 grams of the standard bait and the other dish bottom containing 4 grams of the test product. The test product was ground with a mortar and pestle so that it was similar in size to the corn carrier (unless otherwise noted as whole). After a period of 24 hours the petri dish bottoms were collected and weighed. The dishes had been numbered, weighed and recorded per replication per treatment before being tested. The finished weights were subtracted from the beginning weights. These weights were then recorded and an acceptance ratio (grams candidate bait removed/grams standard bait removed) was calculated for each replication with a mean acceptance ratio calculated per treatment (Table 1). These tests were done both at the Gulfport, MS IFA Laboratory with the red imported fire ant and the Mississippi State IFA Laboratory with the hybrid. Some products were only tested at one site due to availability.

Field tests, conducted June-July 2002, used the most promising commercial products. These products are listed in Table 2. The following test protocol was developed for these tests: 1. Quantity: 1 gram of whole test bait vs standard bait (same as above) in 50mm x 9 mm polystyrene petri dishes. 2. Bait Station: One petri dish bottom with test bait next to one petri dish bottom with standard bait. 3. Location of Bait Stations: Each of 7 treatment bait stations equal distance apart in a circle (10 foot radius) around a mound. Stations were moved at least 1 foot from obvious foraging trails. 4. Number of Replications: The circle of treatments was replicated each time at five separate mounds. These were run on at least five separate dates at five different locations or sets of mounds. The minimum number of replications per treatment was 25. 5. Temperature: Air temperature in shade and soil temperature at 2" depth was recorded at start of test and at finish. 6. Time of Testing: Tests were set out at temperature favorable for ant foraging for approximately one half hour. 7. Termination of Test: Petri dish covers were placed over bottoms to secure ants in dishes. The dishes were systematically collected and returned to the laboratory and placed in a freezer. 8. Collection and Analysis of Data: The ants were counted and recorded by treatment and replication. The t-test was used to compare this paired comparison test of test bait versus standard bait.

This procedure was altered at the hybrid IFA locations first to a test length of one hour. It was then altered to circles with a 5 foot radius and a half hour time length.

RESULTS:

The results of the laboratory tests are presented in Table 1. Corn oil on pregelled corn carrier was by far the most attractive compared to the peanut oil standard for both the red and hybrid imported fire ants. This is however not a commercial product. Many of the products have an acceptance ratio of close to or equal to one (a ratio of 0.75 is generally the minimum acceptable ratio). This indicates that they were similar to or as good as the standard.

The safflower seed coat could not be penetrated by the fire ants and was too big to be removed from the petri dish. The popcorn seed coat caused similar problems in preliminary testing and was impossible to grind with a mortar and pestle.

Table 1. Laboratory Tested Products Versus Standard (Peanut Oil on Pregelled Corn Carrier) with Results Shown as Acceptance Ratios (1 = no difference).

PRODUCT	ACCEPTANCE RATIO	
	RIFA	HYBRID
Vienna Sausage®	1.00	1.00
Spam®	1.00	0.84
Pecan Sandies Cookies®	1.19	0.86
Mini Ritz Crackers®	1.00	0.97
Lay's Orig. Potato Chips®	1.32	0.91
Canned Pressed Ham	-----	1.34
Fritos Corn Chips®	0.21	1.00

Puppy Chow®	0.34	1.00
Peanut Butter	0.70	0.78
Mayonnaise	0.39	0.44
Turtle Food	0.48	0.59
Fish Food	0.66	0.20
Salamander Food	0.36	0.10
Dog Treats	0.50	0.84
Cat Chow®	0.33	1.00
Chezit Crackers®	0.42	0.74
Pork Skins	0.72	0.99
Tostado Corn Chips®	0.34	0.96
Honey	0.14	0.19

All oils were for human consumption except where noted and were presented on the pregelled corn carrier like the standard bait of cooking grade peanut oil.

Corn Oil	1.79	2.49
Soybean Oil	1.00	1.34
Canola Oil	1.00	0.83
Safflower Oil	1.00	1.00
Cod Liver Oil	0.00	0.59
Castor Oil	0.00	0.00
Peanut Oil-Raw	-----	1.00
Olive Oil	1.01	0.65
Soy bean Oil (27%)-Karo Syrup (3%)	-----	1.00
Cottonseed Oil-Once Refined	-----	1.00
Cottonseed Oil-Raw	-----	0.44
Lard	-----	1.00
Macadamia Nuts	0.40	0.78
Almonds	0.73	0.98
Hazel Nuts	0.68	0.27
Chinese Pine Nuts	0.87	0.02
English Walnuts	0.72	1.00
Safflower Seeds-Whole	0.08	0.00
Safflower Seeds-Ground	-----	1.00
Sunflower Seeds-Ground	0.97	0.88
Tast-E-Bait®	0.53	0.83
Artificial Media for Rearing		
Entomophages® -Dry	-----	0.35
AMR Entomophages® -Moist	-----	0.99

The results of the field test for the red imported fire ant are presented in Table 2. This data was analyzed as paired comparisons using the “t” Test. The “t” values show that the Doritos Nacho

Cheesier®, Spam®, Mini Ritz Crackers®, Frito Corn Chips® and Kebbler’s Pecan Sandies® were significantly more attractive than the standard bait.

The field test results for the hybrid imported fire ant are not tabulated. These tests were run 10 times for a total of 50 replications for each of the eight commercial products in Table 2. These were done at 5 different locations on 10 different dates. It was done first at 10 feet from the mound for 30 minutes and then for 1 hour. These were followed by 3 attempts at 5 feet from the mound for 30 minutes. The results in most cases were considered contaminated for the comparison purposes of the testing. This contamination was from competition from at least 8 other species of ants with the Small Black Ant (*Monomorium minimum*) being the most prevalent. There were from 6 to 10 replications of each treatment that were free from competition or “over run”. The “t” values showed that with all treatments tested, the standard bait was the best. Following these June to July (extended into September for the hybrid) field tests, cooler weather tests were attempted in October and November. Unusually heavy rainfall made this work impossible. One test, however, was accomplished for the Red Imported Fire Ant but only 2 of 6 bait comparisons had enough ant numbers to show variability. This means more replications were needed before any conclusions can be made from this data.

In some preliminary experimental trap design comparison tests either Fritos or Ritz were used in paired comparisons of traps. Under these single bait situations both products seemed capable of attracting as many ants as were attracted in the bait test traps. In one test of ten pairs of traps (prototype trap vs open petri dish) run concomitant with the last bait test, the Ritz crackers in the 7 traps with only the hybrid imported fire ant present, attracted a mean trap catch of 168.3 adult workers with a range of 1 to 339 workers at 5 feet from mounds in one half hour. The Small Black Ant had a mean of approximately 113 adults attracted to the Ritz Crackers in the other 13 traps.

Table 2. Field Tests of Eight Commercial Products as Possible Survey Baits for the Red Imported Fire Ant. A Paired Comparison “t” Test ($t=2.064$; $df=24$; $P=0.05$) Was Used to Test Each Product to a Standard Bait.

Bait	t-value	df	Mean Difference	P
Puppy Chow®	1.797	24	4.640	0.085
Lay’s Potato Chips®	1.973	24	10.08	0.060
Doritos Nacho Cheese®	3.059	24	56.960	0.005
Spam®	3.420	23	34.750	0.002
Mini Ritz Crackers®	3.899	24	48.040	0.001
Fritos Corn Chips®	4.002	24	34.640	0.001
Pecan Sandies®	4.075	24	40.120	0.000

DISCUSSION:

Attractiveness is of prime importance in the selection of a bait for use in survey traps. However, you need to consider several other factors such as cost, availability and ease of handling. Any product that has to be cut into little squares and/or leaves the trappers hands greasy becomes labor intensive. This equals higher cost when estimating trapping time with a large scale trapping effort. Spam® fits this category both needing to be cut into quarter inch square pieces and being extremely greasy. Peanut butter, mayonnaise, and honey are others that require cleanup time if traps are to be reused. The Doritos Nacho Cheese® chips need to be broken into smaller pieces as do the Potato Chips and Pecan Sandies®. All three leave messy hands and they crumble into small particles. This crumbling or leaving crumbs around the traps, either from loading the trap at the location or falling from bait loaded traps, could affect the trapping results. The Fritos Original Corn Chips® need minimal breaking and leave few crumbs. This product is probably the most readily available in the field. It is found where ever snack foods are sold in small to large bags. It is being used in some California RIFA Surveys in place of Spam®. The Mini Ritz Crackers® fits well into the California Basket Trap, and will fit easily in many trapping systems, without being broken and weighs slightly under a gram per unit. It has soybean oil listed as its only vegetable oil ingredient while Fritos lists corn and/or sunflower oil. If broken it does leave crumbs. It is readily available but not nearly as common as Fritos. Mini Original Ritz Crackers® are packaged in 312 gram recloseable packages which make them easy to handle. This is the one product that will give as close to a measured dose or bait in each trap as possible for consistency.

Corn oil gave the best results in the laboratory tests. Several of the products listed corn oil as an ingredient, while others included sunflower oil or soybean oil. Many commercial products list simply vegetable oil which can be one of several based on market availability or price. This means that the use of a commercial product such as those tested can always vary as to some of their ingredients. This could have an effect on trap catch numbers or attractiveness of the bait. The need to develop a bait of known composition and quality could be very important for more than consistency. The manufacturers are also not too keen on their nationally advertised products being associated with notorious pest insects.

The results of the field bait tests for the hybrid imported fire ant show that the bait test protocol needs to be changed if further testing is necessary. The protocol worked fine in areas where imported fire ants are numerically dominant. The situation in northern Mississippi is different with the numerous ant species coming to and competing for the baits.

These trials will continue to assess seasonality, and ultimately to pursue development of a standard attractant on an easily handled and distributed substrate.

Literature Cited:

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PROJECT NO: A2P06

PROJECT TITLE: Test and Develop an Imported Fire Ant Survey Trap.

TYPE REPORT: Interim

LEADER/PARTICIPANTS: Robert G. Jones, Ph.D, Timothy Lockley

INTRODUCTION:

Currently, the Federal Imported Fire Ant Quarantine (7CFR301.81) has no recommended survey trap. States and researchers have used a variety of traps with no known consistency of trapping or survey results. Some traps are handmade increasing survey planning and logistic problems. Others are difficult to handle under field conditions. This means increased time needed to set out and collect traps. The collecting, preserving and recording of specimens must be done in the field when it can be done better and faster in a laboratory situation.

The traps being used or have been used include vials and jars of various sizes. The most convenient and economical is a plastic centrifuge tube or vial with an attached lid. Index cards have been used as a substrate or holder for sticky baits, such as peanut butter. The State of California uses a plastic cage to hold the bait (1/4 inch cube of Spam®). In essence these “traps” are containers or substrates for baits. The reason for this is that the foraging ants will carry some of the bait back to the nest leaving a pheromone trail for others to follow. The Survey needs to demonstrate that there are established nests present. To do this catching both major and minor worker IFA or numerous workers produces proof of an established colony. Thus a trap that catches or entraps the foraging ant will not work for this Quarantine Survey.

This is a preliminary report of an effort to develop an IFA Survey trap that is effective, simple and easy to use. Methods used included literature surveys, questioning experienced workers and looking at a variety of containers. Both purchasing and altering a variety of containers was done. These products were then tested and observed in both the laboratory and in the field with IFA. In differing ways, a good survey trap is user friendly to both the subject insect and the surveyor. The following is a list of requirements for a trap that experience, inquiry, observation, and testing have brought forth.

1. Inexpensive because of the large numbers needed and the potential loss rate.
2. Easy to prepare, set out, mark for finding and keep stable on rough ground.
3. Effective in allowing IFA to enter and leave.
4. Holding a bait or attractant that can not be removed but must be fed on thus causing the ants to accumulate and remain as long as possible.
5. Use a clear surface on a portion of the trap so that the contents are visible.
6. The trap must be easy to pickup and the openings quickly shut.
7. The upper surface of the trap needs a reflective surface to prevent overheating inside and contain a program identification logo. Helpful to have some surface of plastic roughened to accept lead pencil markings for location or data information.

8. The traps need to be reasonably small or short and stackable to be placed in carrying containers for field placement including pickup and then in insulated storage chests to preserve specimens.
9. The trap construction must have the ability to withstand temperature changes, such as from the field to a freezer to knock down specimens, and still be reusable.
10. The trap should be multidirectional to increase chance of being entered as opposed to a small vial or jar with a single opening.

A prototype trap has been developed after numerous attempts that fit the above criteria. This prototype is a small petri dish with a tight fitting top. There are eight holes drilled equal distanced around the perimeter. This allows for a simple twist of the top and bottom to close the entrance openings thus capturing the ants. The top of the trap is covered with aluminum foil to prevent heat buildup. As with the baits, the test procedure used in south Mississippi did not work in north Mississippi. The IFA is not numerically dominant in the north and competing ant species confused the test results. Developing a comparative field testing procedure and all field work has been delayed by extreme rainfall during the months of October and November 2002. This followed hot dry weather during July, August and September, 2002.

Copies of the prototype trap have been made and sent to the Project Advisor and the External Collaborator for their comments.

APPENDIX I - LABORATORY BIOASSAY PROCEDURE

PROTOCOL FOR BIOASSAY OF INSECTICIDE TREATED POTTING MEDIA WITH ALATE IFA QUEENS

Introduction: The development of quarantine treatments to prevent artificial spread of imported fire ants (IFA) in nursery stock requires the evaluation of candidate pesticides, dose rates, formulations, etc. The use of a laboratory bioassay procedure for these evaluations provides a rapid and inexpensive means of evaluating the numerous candidates tested each year. Various bioassay procedures have been devised over the years, but the procedure currently used by the USDA, APHIS Imported Fire Ant Laboratory in Gulfport, Mississippi, is described herein. This procedure is a slight modification of the test described by Banks et al., 1964 (J. Econ. Entomol. 57: 298-299).

Collection of test insects: Field collected alate imported fire ant queens are used as the test insect. IFA colonies are opened with a spade and given a cursory examination for the presence of this life stage. Alate queens are seldom, if ever, present in all IFA colonies in a given area. Some colonies will contain only males, others may have few or no reproductive forms present, others may contain both males and queens, while some will contain only alate queens. Seasonal differences in the abundance of queens is quite evident; in the warmer months of the year 50% or more of the colonies in a given area may contain queens. However, in the cooler months, it is not uncommon to find that less than 10% of the colonies checked will contain an abundance of alate queens. Therefore, it is necessary to examine numerous colonies, selecting only those which contain large numbers of alate queens for collection. During winter, ants will often cluster near the surface of the mound facing the sun. Collection during midday on bright, sunny days is highly recommended for winter; whereas the cooler time of day is recommended for hot, dry days of summer. Once a colony (or colonies) has been selected for collection, the entire nest tumulus is shovelled into a 3-5 gallon pail. Pails should be given a liberal dusting with talcum powder on the interior sides to prevent the ants from climbing up the sides of the pail and escaping. Approximately 3-6" head room should be left to prevent escape. An effort should be made to collect as many ants as possible while minimizing the collection of adjacent soil which will contain few ants. Collected colonies are then transported to the laboratory for a 3-5 day acclimation period. The addition of food or water during this short acclimation period is not necessary. Alate queens are collected with forceps after placing a 1-2 liter aliquot of the nest tumulus in a shallow laboratory pan. Again, the use of talc on the sides of containers prevents escape while talced rubber gloves minimizes the number of stings experienced by the collector. The forceps should be used to grasp the queens by the wings in order to prevent mechanical injury. An experienced collector can collect 2-300 queens per hour. It is generally advisable to place collected queens in a 500 cc beaker or other suitable vessel containing moist paper towels prior to being introduced into the test chamber.

Test chambers: Test chambers are 2.5" x 2.5" plastic flower pots which have been equipped with a labstone bottom. Labstone is generally available through dental supply firms such as Patterson Dental Co., 2323 Edenborn Ave., Metairie, Louisiana. The labstone bottom prevents the queens from escaping through the drain holes in the bottom of the pot and also serves as a wick to

absorb moisture from an underlying bed of wet peat moss (see Figure 1). Ants are susceptible to desiccation so humidity/moisture levels must be optimized. Pots should be soaked in water to moisten the labstone prior to placing potting media in the pots. Plastic petri dishes are inverted over the tops of the pots to prevent escape from the top of the test chambers. Prior to placing queens in the test chamber, 50 cc of treated potting media is placed in the bottom of each pot. Due to possible pesticide contamination, test chambers are discarded after use.

Replicates: Each treatment to be evaluated is subdivided into 4 replicates; with one test chamber per replicate. Five alate queens are then introduced into each replicate.

Test interval: All evaluations are based on a 7 day continuous exposure period. i.e., introduced queens remain in the test chambers for 7 days. At this time the contents of each chamber are expelled into a shallow laboratory pan and closely searched for the presence of live IFA alate queens.

Recording of data: Results of each bioassay are entered on the attached data form. Conclusions regarding efficacy and residual activity of the candidate treatments are drawn from this raw data.

Time estimates: The time required to conduct a bioassay will vary greatly, dependent upon a number of factors:

- 1) Availability of queens; supply is primarily influenced by season. More time will be spent collecting queens in winter or during extreme droughts.
- 2) Number of treatments to be evaluated; e.g., if only a single treatment and an untreated check are to be evaluated only 40 queens/month are needed. Conversely, a test involving 4 insecticides at 3 rates of application (12 treatments + untreated check) will require 260 queens monthly for the duration of the test.

Duration of the trial: A successful preplant incorporated treatment for nursery potting soil must provide a minimum of 12-18 months residual activity in order to conform with normal agronomic practices of the nursery industry. Since some plants may be held for longer periods of time prior to sale, a 24-36 month certification period (residual activity) would be ideal. Therefore, most initial or preliminary trials with a given candidate treatment are scheduled for 18 months.